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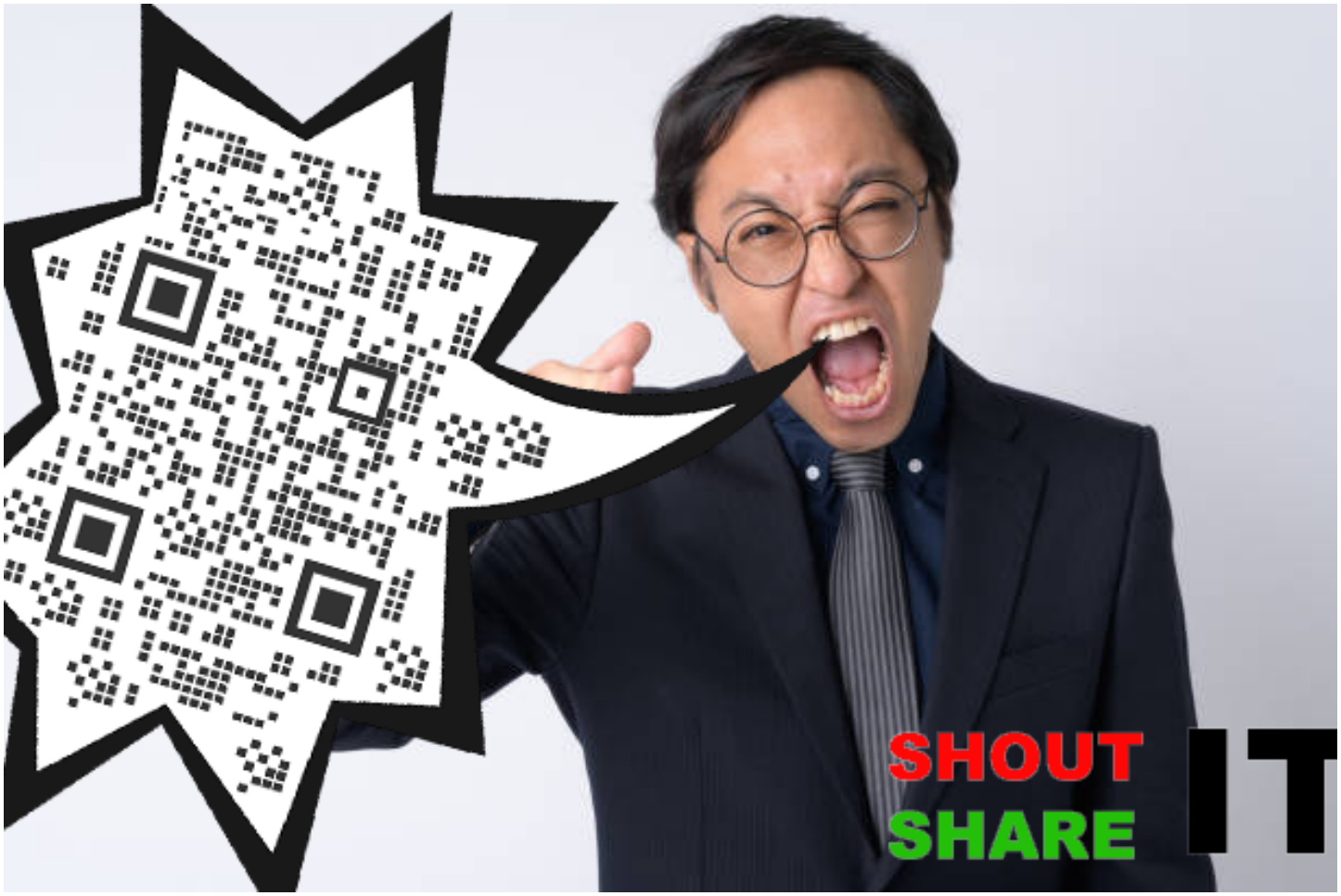
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A Bibliography

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Biotechnology of Algae:

A Bibliography

A selected bibliography of research and product development using genetic engineering and molecular biology techniques and species of fresh water and marine algae.

by Virginia Stone
and Robert D. Warmbrodt

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Preface

The field of biotechnology continues to expand rapidly with innovative research, new technologies, and the development of beneficial products for animals and humans. The many disciplines using genetic engineering and molecular biology techniques attest to the suggestion that biotechnology is truly a multidisciplinary field. In addition, the species used as investigative tools are as wide ranging and varied as the biological sciences themselves. For example, in agriculture, virtually all of the major plant commodities and animal species such as swine, cattle, sheep, and fish are the subject of biotechnology research, non-photosynthetic microorganisms such as *Bacillus thuringiensis*, yeasts, and Rhizobium spp. play a critical role in biotechnology and even numerous species of fresh and marine water algae contribute to both basic and applied research and product development in biotechnology.

The use of algae in biotechnology research and in the biotechnology industry is significant. Algae play critical roles as bioreactors for the production of food, chemicals, and fuels. They are becoming extremely important in the development of solar energy technology and in biodegradation and bioremediation programs, and their importance in the ever-expanding domestic and international aquaculture industry cannot be over-emphasized.

Because of the economic importance of this diverse group of organisms, NAL's Biotechnology Information Center, in conjunction with its Aquaculture Information Center, has compiled this bibliography of basic and applied research on algae and biotechnology. The citations listed herein represent research that was selected for its creativity, innovation, and timeliness. The bibliography will be invaluable to researchers, industry representatives, government officials, environmental groups, the interested public, and others interested in algae and biotechnology.

This bibliography has been sub-divided into several sections representing the major efforts in algal biotechnology research. The first section represents literature of a general nature followed by sections on the specific topics of culture, gene expression and sequencing information, products and product development, and finally, bioremediation and biodegradation. An author index follows the bibliographic text.

The citations included in this bibliography were taken from the NAL AGRICOLA database and from BIOSIS Previews. In addition to the title, author, source, and, where available, an abstract, each citation also includes key descriptors and the NAL Call Number if the material is part of the NAL collection.

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ROBERT D. WARMBRODT
COORDINATOR, BIOTECHNOLOGY INFORMATION CENTER
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Availability of Cited Documents

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Biotechnology of Algae: A Bibliography

General Interest

1
Introduction to Applied Phycology
Akatsuka, I.
Source: Academic Publishing, The Hague, Netherlands, 1990, 683 pp.
Descriptors: book; commercial uses; genetic engineering; toxicity; tissue culture
Abstract:

This reference work presents the latest research into the commercial use of algae. Techniques and practical applications are described. The topics covered include biotechnology, genetic engineering, tissue culture, pollution and toxicity. The text is supplemented by diagrams, graphs, tables, chemical compounds diagrams, photographs, an author index and a subject index.

2
Algal and Cyanobacterial Biotechnology
Cresswell, R.C.; Rees, T.A.V.; Shah, N., editors
Source: Wiley, New York, 1989, 341 pp.
Descriptors: algae-biotechnology; cyanobacteria-biotechnology
DNAL Call No.: TP248.27.A46A44

3
Seaweed Biotechnology Current Status and Future Prospects
Evans, L.V. and Butler, D.M.
Source: PROCEEDINGS OF THE PHYTOCHEMICAL SOCIETY OF EUROPE, vol. 28. Biochemistry of the Algae and Cyanobacteria; Aberystwyth, Wales, UK, April 1987. Rogers, L.J. and Gallon, J.R., editors. Oxford University Press, New York, 1989, pp. 335-350.
Descriptors: agar; alginic acid; carrageenan; biotechnology; protoplasts; seaweeds; reviews; in vitro culture; seaweed products; enteromorpha; porphyra; gracilaria; fucus

4
Yeasts Molds and Algae
Jacobson, G.K. and Jolly, S.O.
Source: BIOTECHNOLOGY: A COMPREHENSIVE TREATISE, vol. 7B. Gene Technology. VCH Publishers, Inc., New York, 1989, pp. 279-314.
Descriptors: review; food; bioconversion; baking industry; brewing industry; dairy industry; biomass conversion; wastewater treatment; soil fertilizers; fine chemicals; genetic engineering; biotechnology
DNAL Call No.: QR53.B52

5
The Biotechnology of Microalgae and Cyanobacteria
Kerby, N.W. and Stewart, W.D.P.
Source: PROCEEDINGS OF THE PHYTOCHEMICAL SOCIETY OF EUROPE, vol. 28. Biochemistry of the Algae and Cyanobacteria; Aberystwyth, Wales, UK, April 1987. Rogers, L.J. and Gallon, J.R., editors. Oxford University Press, New York, 1989, pp. 319-334.

Descriptors: biomass; ammonia; amino acids; pigments; toxins; animal; cell growth; stimulants; lipids; fatty acids; antibiotics; plant growth stimulants; restriction endonucleases
DNAL Call No.: QK898.T4E26

6
Comparative Physiology and Biochemistry of Chlorella Species as the Basis for their Taxonomy and for their Utilization in Research and Biotechnology
Kessler, E.
Source: PHYCOTALK, vol. 1. Kumar, H.D., editor. Rastogi and Co., Subhash Bazar, Meerut, India, 1989, pp. 141-154.
Descriptors: Chlorella-fusca; Chlorella-vulgaris; Chlorella-sorokiniana; Chlorella-saccharophila; Chlorella-zofingiensis; Chlorella-minutissima; Chlorella-homosphaera; Chlorella-kessleri; Chlorella-luteoviridis; Chlorella-protothecoides

7
Phycotechnology Yesterday Today and Tomorrow Maybe
Lewin, R.A.
Source: BULL MAR SCI 47(1):256-257 (1990).
Descriptors: abstract; spirulina; dunaliella; protozoa contamination

8
Bioconversion of Seaweeds
Morand, P.; Carpentier, B.; Charlier, R.H.; Maze, J.; Orlandini, M.; Plunkett, B.A.; De Waart, J.
Source: SEAWEED RESOURCES IN EUROPE: USES AND POTENTIAL. Guiry, M.D. and Blunden, G., editors. John Wiley and Sons, Inc., Somerset, New Jersey, 1991, pp. 95-148.
Descriptors: marine algae; seaweed cultivation; fuel; biomass; bioengineering; Europe
DNAL Call No.: SH390.7.S44

9
Permeabilized Cyanobacteria: A Model System for Photosynthetic and Biotechnological Studies
Papageorgiou, G.C.
Source: NATO ASI SERIES: SERIES A: LIFE SCIENCES 168:449-467 (1989).
Descriptors: cyanobacteria; biotechnology; permeability; photosynthesis; ultrastructure; electron microscopy; literature reviews
DNAL Call No.: QH301.N32

10
Seaweeds and Biotechnology Inseparable Companions
Renn, D.W.
Source: HYDROBIOLOGIA 204-205(0):7-14 (1990).
Descriptors: polysaccharides; algin; carrageenan; agar; agarose; separation techniques
DNAL Call No.: 410 H992

11
Recent Advances in Microalgal Biotechnology
Vonshak, A.
Source: BIOTECHNOLOGY ADVANCES 8(4):709-728 (1990).
Descriptors: spirulina; dunaliella; chlorella haematococcus; algae; biotechnology industry; biomass conversion
DNAL Call No.: TP248.2.B562

Culture

12
The Biotechnology of Cultivating the Halotolerant Alga Dunaliella
Ben-Amotz, A.; Avron, M.
Source: TRENDS IN BIOTECHNOLOGY 8(5):121-126 (1990).
Descriptors: dunaliella; biotechnology; algae culture; plant products; salt tolerance
DNAL Call No.: TA166.T72

13
Effects of Salinity Increase on Carotenoid Accumulation in the Green Alga Dunaliella-salina
Borowitzka, M.A.; Borowitzka, L.J.; Kessly, D.
Source: JOURNAL OF APPLIED PHYCOLOGY 2(2):111-120 (1990).
Descriptors: food industry; lutein; beta carotene; alpha carotene; shock; osmotic stress; biotechnology
DNAL Call No.: QK564.J68
Abstract:

The effect of sudden salinity increases on the kinetics of growth and carotenogenesis was studied in three geographically diverse isolates of Dunaliella salina. A sudden increase in salinity results in a lag phase in growth and the length of this lag phase is dependent on the final salinity and the magnitude of the salinity change (no lag at 10-15% w/v NaCl, 4-day lag at 30% NaCl). There is also a lag before an increase in the total carotenoid content can be measured following the salinity up-shock, and the length of the lag depends largely on the initial salinity and the magnitude of the salinity up-shock, whereas the rate of carotenogenesis and the final carotenoid content reached depend on the final salinity. The increase in total carotenoid content is mainly due to .beta.-carotene. Following the salinity up-shock (especially from 10% to 20% NaCl) the proportion of lutein as a percentage of total carotenoids decreases, whereas zeaxanthin increases. This suggests that the pathway synthesising lutein is more sensitive to salt or osmotic stress and is inhibited at higher salinities thus leading to .beta.-carotene formation. The proportion of .alpha.-carotene does not change.

14
On-Line Optimization of Biotechnological Processes I. Application to Open Algal Pond
Guterman, H. and Ben-Yaakov, S.
Source: BIOTECHNOLOGY AND BIOENGINEERING 35(4):417-426 (1990).
Descriptors: algae; biotechnology; production; ponds; mathematical models
DNAL Call No.: 381 J8224
Abstract:

A new on-line optimization and control procedure applicable to biotechnological systems for which a precise mathematical model is unavailable has been developed and tested. The proposed approach is based on an on-line search for optimum conditions by an automatic system using a modified simplex algorithm to which several features have been added to permit real time operation. The simplex algorithm is the upper level of a hierarchical software package in which the other levels are cost evaluation, control, data acquisition, and signal processing. The optimization method was tested in a laboratory minipond for the cultivation of Spirulina platensis. The controlled parameters were light intensity, optical density, pH, and temperature. The proposed optimization method can be applied to other biological processes provided that the pertinent variables can be measured and controlled and the cost function can be defined mathematically.

15
The Mass Culture of Dunaliella-viridis volvocales chlorophyta for Oxygenated Carotenoids Laboratory and Pilot Plant Studies
Moulton, T.P. and Burford, M.A.
Source: HYDROBIOLOGIA 204-205(0):401-408 (1990).
Descriptors: dunaliella-salina; beta carotene; biotechnology
DNAL Call No.: 410 H992

16
Effects of Light Intensity on the Growth Rate of the Red Alga Porphyridium-cruentum
Sada, E.; Katoh, S.; Kheirolomoon, A.; Yokoi, H.
Source: JOURNAL OF FERMENTATION AND BIOENGINEERING 67(2):135-137 (1989).
Descriptors: batch fermentation; continuous fermentation; arachidonic acid; yield; biotechnology industry; pharmaceuticals
DNAL Call No.: QP601.A1J6
Abstract:

The red alga, Porphyridium cruentum, which is one of the potential sources of arachidonic acid, was cultured in batch and continuous vessels. The growth rates in batch cultures were correlated to the mean light intensity in the vessels, and the cell concentrations in continuous cultures were estimated by those results. The yield of arachidonic acid was about 1.2 g per 1012 cell at cell concentrations ranging from 0.5 to 1.5 .times. 1010 cell/l and independent of the mean light intensity.

17
Macroalgal Strain Selection and Improvement in Japan
Saga, N.
Source: BULL MAR SCI 47(1):260 (1990).
Descriptors: abstract; food; energy source; chemical production; breeding; cultivation; biotechnology

18
Mass Culture of Spirulina-fusiformis and its Nutritional and Toxicological Evaluation
Seshadri, C.V.
Source: FOOD BIOTECHNOLOGY 4(1):607 (1990).
Descriptors: abstract; algae; food; protein; vitamin bioavailability; mineral bioavailability; biotechnology
DNAL Call No.: TP248.65.F66F66

19
Growth Chemical Composition of Cyanobacteria Spirulina-maxima in Batch Cultures
Tadros, M.; Tadros, S.; Smith, W.; Mbuthia, P.; Joseph, B.
Source: ABSTR ANNU MEET AM SOC MICROBIOL 90(0):233.
DNAL Call No.: 448.39.SO12A

20
Porphyra Cell Cultures Isolation Growth and Polysaccharide Production
Tait, M.I.; Milne, A.M.; Grant, D.; Somers, J.A.; Staples, J.; Long, W.F.; Williamson, F.B.; Wilson, S.B.
Source: JOURNAL OF APPLIED PHYCOLOGY 2(1):63-70 (1990).
Descriptors: bioengineering; nutrients
DNAL Call No.: QK564.J68

Abstract:

A range of cell lines was isolated from Porphyra umbilicalis L. (Rhodophyta) tissue using a variety of methods, the most successful involving exposure to a limpet acetone powder enzyme extract for 24 h, homogenisation and filtration through a series of polyester meshes. All established lines grew as 0.1-5 mm diameter aggregates in liquid culture; most were stable and have been grown in shake-flask or air-lift culture for periods in excess of 1 yr without reverting to the foliose growth form. An investigation of the medium used to grow these lines indicated that it was not nitrogen-deficient and that the sodium chloride concentration was optimal. The addition of an organic buffer increased the final cell yield. None of these cell lines grew heterotrophically in medium supplemented with a range of fixed carbon sources. The infrared spectra of polysaccharides isolated from Porphyra aggregates and from tissue grown under identical conditions indicated that the structures of the two isolates were analogous.

21

Absorption of Carbon Dioxide in Algal Mass Culture Systems a Different Characterization Approach

Talbot, P.; Gortares, M.P.; Lencki, R.W.; De la Noue, J.

Source: BIOTECHNOLOGY AND BIOENGINEERING 37(9):834-842

Descriptors: algae; microorganism; biotechnology industry; mass transfer; kinetics; photosynthesis; bioreactor

DNAL Call No.: 381 J8224

Abstract:

For the characterization of CO2 absorption in aerated microalgal culture systems, a different approach based on KLa(02) determination and transformation was studied. To confirm the validity of this method, the influence of reactions between CO2 and compounds (OH-, H2O, and NH3) present in the culture medium upon the absorption mechanism was evaluated under different physical and chemical culture conditions. Under these conditions, knowledge of the relative magnitudes of the diffusion and reaction kinetics permitted the evaluation of their relative importance. For the determination of the parameters required for the calculation of the CO2 absorption constant, empirical correlation calculations for KLO and a were used that had been previously verified with experimental data for O2 absorption. Since, for the conditions studied, the absorption rate was shown to be independent of the chemical reactions taking place in the liquid phase, the KLa for CO2 could be directly related to the KLa for O2 by a simple factor that took into account the difference in aqueous diffusivity of the two gases. Thus, using methods developed for determining O2 absorption in gas-liquid contactors, it is possible to adequately characterize CO2 absorption for laboratory and pilot scale algal production systems.

22

Effect of Low-Dose Ultrasonic Treatment on Growth Rates and Biomass Yield of Anabaena-flos-aquae and Selenastrum-capricornutum

Thomas, B.J.; McIntosh, D.; Taylor, S.R.; Francko, D.A.; Ownby, J.

Source: BIOTECHNOLOGY TECHNIQUES 3(6):389-392 (1989).

Descriptors: chlorophyta; biomass production; growth rate; yields; ultrasonic treatment; cell culture; biotechnology

DNAL Call No.: TP248.24.B55

Abstract:

Major project tasks included assembly of an ultrasonic treatment array and measurement of the cell culture growth rate as a function of ultrasonic frequency, and ultrasonic power level and dosage. Growth rates for Anabaena flos aquae were increased with both single or multiple ultrasonic dosages and were over and above that obtained with vigorous

mechanical stirring. Selenastrum capricornutum growth rates were decreased by ultrasonic treatment. The results were also shown to be independent of the degree of cell agglomeration. Collectively, the data support the conclusion that low-dose, short duration ultrasonic treatment induces changes in culture growth rates in both algal species examined.

23
Phototropism in Dunaliella and its Application in a Harvesting Device
Toha, J.; Soto, M.A.; Contreras, S.
Source: BIOTECHNOLOGY TECHNIQUES 4(5):321-324 (1990).
Descriptors: algae; methods; equipment; biotechnology; large scale culture
DNAL Call No.: TP248.24.B55
Abstract:

The influence of wavelength, light intensity and algal concentration on the phototropism of Dunaliella sp. is described. A practical device for harvesting the alga, based in this effect is shown.

24
Biotechnology Studies on the Breeding of Porphyra-yezoensis ueda
Wang, S.; Zhou, Y.; He, P.
Source: JOURNAL OF PHYCOLOGY 27 (3 suppl.):75 (1991).
Descriptors: abstract; somatic cells; growth; development; spore yield; mariculture
DNAL Call No.: QK564.J6

Gene Expression and Sequencing Studies

25
Characterization of the IS895 Family of Insertion Sequences from the Cyanobacterium Anabaena sp. strain PCC 7120
Alam, J.; Vrba, J.M.; Cai, Y.; Martin, J.A.; Weislo, L.J.; Curtis, S.E.
Source: JOURNAL OF BACTERIOLOGY 173(18):5778-5783 (1991).
Descriptors: anabaena; strains; transposable elements; nucleotide sequences; amino acid sequences
DNAL Call No.: 448.3 J82
Abstract:

A family of repetitive elements from the cyanobacterium Anabaena sp. strain PCC 7120 was identified through the proximity of one element to the psbA1 gene. Four members of this seven-member family were isolated and shown to have structures characteristic of bacterial insertion sequences. Each element is approximately 1,200 bp in length, is delimited by a 30-bp inverted repeat, and contains two open reading frames in tandem on the same DNA strand. The four copies differ from each other by small insertions or deletions, some of which alter the open reading frames. By using a system designed to trap insertion elements, one of the elements, denoted IS895, was shown to be mobile. The target site was not duplicated upon insertion of the element. Two other filamentous cyanobacterial strains were also found to contain sequences homologous to IS895.

26
Evolution of the Rubisco Operon from Prokaryotes to Algae: Structure and Analysis of the rbcS Gene of the Brown Alga Pylaiella littoralis
Assali, N.E.; Martin, W.F.; Sommerville, C.C.; Loiseaux-de Goer, S.

Source: PLANT MOLECULAR BIOLOGY: AN INTERNATIONAL JOURNAL ON MOLECULAR BIOLOGY, BIOCHEMISTRY AND GENETIC ENGINEERING 17(4):853-863 (1991).
Descriptors: phaeophyta; genes; ribulose-bisphosphate carboxylase; nucleotide sequences; amino acid sequences; plastids; genomes; phylogeny; restriction mapping; transcription
DNAL Call No.: QK710.P62
Abstract:

The rbcS gene coding for the small subunit of ribulose-1,5-bisphosphate carboxylase/oxygenase (Rubisco) of the brown alga Pylaiella littoralis is located within the plastid genome and is transcribed as a single polycistronic mRNA with the gene for the large subunit of Rubisco, rbcL. The structure of the Rubisco operon from P. littoralis was determined. Molecular phylogenies for rbcS and rbcL with a wide range of prokaryotes and eukaryotes were constructed which are congruent with recent evidence for polyphyletic plastid origins. Both rbcL and rbcS of the beta-purple bacterium Alcaligenes eutrophus clearly cluster with the rhodophyte and chromophyte proteins. The data suggest that the Rubisco operons of red algal and chromophytic plastids derive from beta-purple eubacterial antecedents, rather than the cyanobacterial lineage of eubacteria from which other of their genes derive. This implies a lateral transfer of Rubisco genes from beta-purple eubacterial ancestors to the cyanobacterial ancestor of rhodophyte and chromophyte plastids.

27
Different Fates of the Chloroplast tufA Gene Following its Transfer to the Nucleus in Green Algae
Baldauf, S.L.; Manhart, J.R.; Palmer, J.D.
Source: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA 87(14):5317-5321 (1990).
Descriptors: algae; chloroplasts; dna; evolution; genetic code; genetics
DNAL Call No.: 500 N21P

28
Characterization of an Insertion Sequence (IS891) of Novel Structure from the Cyanobacterium Anabaena sp. Strain M-131
Bancroft, I. and Wolk, C.P.
Source: JOURNAL OF BACTERIOLOGY 171(11):5949-5954 (1989).
Descriptors: anabaena; strains; nucleotide sequence; characterization
DNAL Call No.: 448.3 J82
Abstract:

When recombinant plasmids that here transferred to the cyanobacterium Anabaena sp. strain M-131 were transferred back to Escherichia coli, some of the transformants contained inserts. One of the insertion sequences (ISs) as characterized by sequencing. This 1,351-base-pair IS contained an open reading frame that was capable of encoding a peptide of 310 amino acids and had terminal sequences with distinctive structures, but it lacked terminal inverted repeats and did not duplicate target DNA upon insertion. The element bore no significant sequence homology to any sequence stored in the GenBank data base. Restriction analysis of the genomes of Anabaena sp. strain M-131 and Anabaena sp. strain PCC 7120 showed those strains to be closely related. Sequences homologous to the IS element were also present in the DNA of Anabaena strain PCC 7120, but the copy numbers and chromosomal locations or such sequences differed in the two strains. The largest visualized plasmid was 425 kilobases (kb) in M-131 and 410 kb in PCC 7120; at least the former plasmid contained multiple copies of the element, as did a 115-kb plasmid in M-131.

29
Studies on Chlamydomonas Chloroplast Transformation: Foreign DNA can be Stably Maintained in the Chromosome
Blowers, A.D.; Bogorad, L.; Shark, K.B.; Sanford, J.C.
Source: THE PLANT CELL 1(1):123-132 (1989).
Descriptors: chlamydomonas reinhardtii; chloroplast genetics; genetic transformation; homologous recombination; dna; dna hybridization; repetitive dna; restriction mapping; messenger rna; northern blotting; gene expression; chimeras
DNAL Call No.: QK725.P532

30
Transcriptional Analysis of Endogenous and Foreign Genes in Chloroplast Transformants of Chlamydomonas
Blowers, A.D.; Ellmore, G.S.; Klein, U.; Bogorad, L.
Source: THE PLANT CELL 2(11):1059-1070 (1990).
Descriptors: chlamydomonas reinhardtii; chloroplast genetics; genetic transformation; chloroplasts; genes; beta-glucuronidase; reporter genes; transcription; promoters; genetic regulation; gene expression; deletions; mutagenesis; nucleotide sequences; homologous recombination
DNAL Call No.: QK725.P532

31
Characterization of Insertion Sequence IS892 and Related Elements from the Cyanobacterium Anabaena sp. Strain PCC 7120
Cai, Y.
Source: JOURNAL OF BACTERIOLOGY 173(18):5771-5777 (1991).
Descriptors: anabaena; strains; transposable elements; nucleotide sequences; amino acid sequences
DNAL Call No.: 448.3 J82
Abstract:

IS892, one of the several insertion sequence (IS) elements discovered in Anabaena sp. strain PCC 7120 (Y. Cai and C.P. Wolk, J. Bacteriol. 172:3138-3145, 1990), is 1,675 bp with 24-bp near-perfect inverted terminal repeats and has two open reading frames (ORFs) that could code for proteins of 233 and 137 amino acids. Upon insertion into target sites, this IS generates an 8-bp directly repeated target duplication. A 32-bp sequence in the region between ORF1 and ORF2 is similar to the sequence of the inverted termini. Similar inverted repeats are found within each of those three segments, and the sequences of these repeats bear some similarity to the 11-bp direct repeats flanking the 11-kb insertion interrupting the nifD gene of this strain (J.W. Golden, S.J. Robinson, and R. Haselkorn, Nature [London] 314:419-423, 1985). A sequence similar to that of a binding site for the Escherichia coli integration host factor is found about 120 bp from the left end of IS892. Partial nucleotide sequences of active IS elements IS892N and IS892T, members of the IS892 family from the same Anabaena strain, were shown to be very similar to the sequence of IS892.

32
Use of a Conditionally Lethal Gene in Anabaena sp. Strain PCC 7120 to Select for Double Recombinants and to Entrap Insertion Sequences
Cai, Y. and Wolk, C.P.
Source: JOURNAL OF BACTERIOLOGY 172 (6):3138-3145 (1990).
Descriptors: anabaena; strains; lethals; recombination
DNAL Call No.: 448.3 J82
Abstract:

Use of the sacB gene (J.L. Ried and A. Collmer, Gene 57:239-246, 1987) provides a

simple, effective, positive selection for double recombinants in Anabaena sp. strain PCC 7120, a filamentous cyanobacterium. This gene, which encodes the secretory levansucrase of Bacillus subtilis, was inserted into the vector portion of a suicide plasmid bearing a mutant version of a chromosomal gene. Cells of colonies in which such a plasmid had integrated into the Anabaena chromosome through single recombination were plated on solid medium containing 5% sucrose. Under this condition, the presence of the sacB gene is lethal. A small fraction of the cells from initially sucrose-sensitive colonies became sucrose resistant; the majority of these sucrose-resistant derivatives had undergone a second recombinational event in which the sacB-containing vector had been lost and the wild-type form of the chromosomal gene had been replaced by the mutant form. By the use of this technique, we mutated two selected genes in the chromosome of Anabaena sp. strain PCC 7120. The conditionally lethal nature of the sacB gene was also used to detect insertion sequences from this Anabaena strain. Sucrose-resistant colonies derived from cells bearing a sacB-containing autonomously replicating plasmid were analyzed. Five different, presumed insertion sequences were found to have inserted into the sacB gene of the plasmids in these colonies. One of them, denoted IS892, was characterized by physical mapping. It is 1.7 kilobases in size and is present in at least five copies in the genome of Anabaena sp. strain PCC 1720.

33

Codon Usage in Higher Plants, Green Algae, and Cyanobacteria
Campbell, W.H. and Gowri, G.
Source: PLANT PHYSIOLOGY 92(1):1-11 (1990).
Descriptors: cyanobacteria; chlorophyceae; plant breeding; protein synthesis; genetic code; codon; genome analysis
DNAL Call No.: 450 P692
Abstract:

Codon usage is the selective and nonrandom use of synonymous codons by an organism to encode the amino acids in the genes for its proteins. During the last few years, a large number of plant genes have been cloned and sequenced, which now permits a meaningful comparison of codon usage in higher plants, algae, and cyanobacteria. For the nuclear and organellar genes of these organisms, a small set of preferred codons are used for encoding proteins. Codon usage is different for each genome type with the variation mainly occurring in choices between codons ending in cytidine (C) or guanosine (G) versus those ending in adenosine (A) or uridine (U). For organellar genomes, chloroplastic and mitochondrial proteins are encoded mainly with codons ending in A or U. In most cyanobacteria and the nuclei of green algae, proteins are encoded preferentially with codons ending in C or G. Although only a few nuclear genes of higher plants have been sequenced, a clear distinction between Magnoliopsida (dicot) and Liliopsida (monocot) codon usage is evident. Dicot genes use a set of 44 preferred codons with a slight preference for codons ending in A or U. Monocot codon usage is more restricted with an average of 38 codons preferred, which are predominantly those ending in C or G. But two classes of genes can be recognized in monocots. One set of monocot genes uses codons similar to those in dicots, while the other genes are highly biased toward codons ending in C or G with a pattern similar to nuclear genes of green algae. Codon usage is discussed in relation to evolution of plants and prospects for intergenic transfer of particular genes.

34

Expression of the Mosquitocidal-Protein Genes of Bacillus thuringiensis subsp. israelensis and the Herbicide-Resistance Gene Bar in Synechocystis PCC6803
Chungjatupornchai, W.

Source: CURRENT MICROBIOLOGY 21(5):283-288 (1990).
Descriptors: bacillus thuringiensis subsp. israelensis; cyanobacteria; bacterial proteins; genes; gene transfer; genetic transformation; promoters; marker genes; gene expression; insecticidal action; culicidae; biological control agents; biological control; herbicide resistance; genetic models
DNAL Call No.: QR1.C78

35
Group II Twintron: an Intron within an Intron in a Chloroplast Cytochrome b-559 Gene
Copertino, D.W. and Hallick, R.B.
Source: THE EMBO JOURNAL-EUROPEAN MOLECULAR BIOLOGY ORGANIZATION 10(2):433-442 (1991).
Descriptors: euglena; chloroplasts; cytochromes; genetic code; photosystem ii; introns; transposable elements; gene mapping
DNAL Call No.: QH506.E46
Abstract:

The psbF gene of chloroplast DNAs encodes the beta-subunit of cytochrome b-559 of the photosystem II reaction center. The psbF locus of Euglena gracilis chloroplast DNA has an unusual 1042 nt group H intron that appears to be formed from the insertion of one group II intron into structural domain V of a second group II intron. Using both direct primer extension cDNA sequencing and cDNA cloning and sequencing, we have determined that a 618 nt internal intron is first excised from the 1042 nt intron of psbF pre-mRNA, resulting in a partially spliced pre-mRNA containing a 424 nt group II intron with a spliced domain V. The 424 nt intron is then removed to yield the mature psbF mRNA. Therefore, the 1042 nt intron of psbF is a group II intron within another group II intron. We use the term 'twintron' to define this new type of genetic element. Intermediates in the splicing pathway were detected by northern hybridization. Splicing of both the internal and external introns occurs via lariat intermediates. Twintron splicing was found to proceed by a sequential pathway, the internal intron being removed prior to the excision of the external intron. A possible mechanism for twintron formation by intron transposition is discussed.

36
Amplified Expression of Ribulose Bisphosphate Carboxylase/Oxygenase inpBR322-Transformants of Anacystis nidulans
Daniell, H.; Torres-Ruiz, J.A.; Inamdar, A.; McFadden, B.A.
Source: ARCHIVES OF MICROBIOLOGY 151(1):59-64 (1989).
Descriptors: anacystis nidulans; enzyme activity; ribulose-bisphosphate carboxylase; plasmids; recombination; chromosomes
DNAL Call No.: 442.8 AR26

37
A Transposon with an Unusual LTR Arrangement from Chlamydomonas reinhardtii Contains an Internal Tandem Array of 76 bp Repeats
Day, A. and Rochaix, J.D.
Source: NUCLEIC ACIDS RESEARCH 19(6):1259-1266 (1991).
Descriptors: chlamydomonas reinhardtii; transposable elements; nucleotide sequences
DNAL Call No.: QD341.A2N8
Abstract:

TOC1, a transposable element from Chlamydomonas reinhardtii, is 5662 bases long. The 217 and 237 base long terminal repeat sequences of TOC1 are unusually arranged around the 4600 and 123 base unique regions: [217]-4600-[237][217]-123-[237]. Although TOC1 contains long terminal repeats and most TOC1 elements are complete, features shared

with virus-like retroposons, its unique 4600 base region is more similar to the structure of the L1 family of non-virus retroposons: first, 11 3/4 tandemly repeated copies of a 76 base repeat are found 813 bases from the left end of TOC1, and second using the universal genetic code large open reading frames were not found in TOC1. The relationship between TOC1, virus-like retroposons and the L1 family of non-virus retroposons is unclear and may be very distant since only poor similarity was found between the TOC1 encoded ORFs and retrovirus polypeptides. The length of the tandem array of 76 base repeat sequences was conserved in most TOC1 elements and solo 76 base repeat sequences were not found outside TOC1 elements in the C. reinhardtii genome. Nucleotide substitutions allow all copies of the 76 base repeat to be distinguished from one another.

38

Structure and Inheritance of Sense and Anti-sense Transcripts from a Transposon in the Green Alga Chlamydomonas reinhardtii

Day, A. and Rochaix, J.D.

Source: JOURNAL OF MOLECULAR BIOLOGY 218(2):273-291 (1991).

Descriptors: chlamydomonas reinhardtii; strains; transposable elements; transcription; patterns; antisense rna; inheritance; progeny; strain differences; molecular mapping; genetic analysis

DNAL Call No.: 442.8 J8224

Abstract:

We have studied the transcription pattern of a 5700 base-pair transposon (TOC1) in Chlamydomonas reinhardtii. Northern blotting and nuclease S1 protection experiments define three classes of major TOC1 RNAs that accumulate to different levels in a number of strains and segregate independently in the progeny of crosses: class 1 RNAs are unstable near full-length sense transcripts whose 5' end maps to the left 217 base-pair repeat of TOC1, class 2 and class 3 RNAs are large, discrete chimaeric transcripts containing full-length sense (class 2) and anti-sense (class 3) copies of TOC1. Sequence motifs common to the 5' non-transcribed regions of C. reinhardtii genes were found upstream from the putative initiation site of class 1 transcripts. A functional polyadenylation site was located in the far-right 237 base-pair repeat of TOC1. Class 1 TOC1 transcripts are initiated, and probably terminated, within the terminal repeats of TOC1 and may represent retrotransposition intermediates. Class 2 and 3 TOC1 transcripts co-segregate with specific TOC1 elements identified on Southern blots. The loci that control the production of high levels of class 1 transcripts could correspond to specific TOC1 elements, i.e. only a few TOC1 elements are transcribed, or to a regulatory locus. The accumulation of an 11,500 to 12,000 base sense transcript (class 2) is reduced two- to fourfold by the presence of a 9500 to 9700 base anti-sense transcript (class 3). In contrast, the accumulation of the 5' ends of class 1 transcripts are unaffected by the anti-sense TOC1 transcript.

39

Genetic Analysis of a 9 kDa Phycocyanin-Associated Linker Polypeptide

De Lorimier, R.; Bryant, D.A.; Stevens, S.E. Jr.

Source: BIOCHIMICA ET BIOPHYSICA ACTA: INTERNATIONAL JOURNAL OF BIOCHEMISTRY AND BIOPHYSICS 1019(1):29-41 (1990).

Descriptors: synechococcus; strains; gene mapping; genetic analysis; genetic code; molecular genetics; mutations; nucleotide sequences; polypeptides; amino acid sequences; cyanin

DNAL Call No.: 381 B522

Abstract:

The gene encoding LR9, a 9kDa phycocyanin-associated linker polypeptide, was cloned from the cyanobacterium Synechococcus sp. PCC 7002 (Agmenellum quadruplicatum PR-

6). This gene, termed cpcD was located immediately 3' to cpcC, a gene which encodes another phycocyanin-associated linker, LR33. Mutation of cpcD by insertion led to the loss of LR9 as the only detectable change in phycobilisome composition. Cells and isolated phycobilisomes from the cpcD- strain did not detectably differ from the wild-type in absorption or steady-state fluorescence emission. Purified phycobilisomes from the wild-type and cpcD- strains were compared by electron microscopy. The number of phycocyanin discs in the rod substructures of the mutant was more variable than in the wild-type. Hence, one function of LR9 may be to minimize the heterogeneity of rod length, possibly by binding to the core-distal face of phycocyanin-LR33 complexes to prevent the tandem joining of such units. A mutant in which cpcD and cpcC-cpcD intergenic sequences are deleted shows a partial loss of LR33. Inverted repeats in this intergenic region may be required for optimal stability of the cpcC transcript.

40

Molecular and Biophysical Analysis of Herbicide-Resistant Mutants of Chlamydomonas reinhardtii: Structure-Function Relationship of the Photosystem II D1 Polypeptide

Erickson, J.M.; Pfister, K.; Rahire, M.; Togasaki, R.K.; Mets, L.; Rochaix, J.D.

Source: THE PLANT CELL 1(3):361-371 (1989).

Descriptors: chlamydomonas reinhardtii; genes; photosystem ii; polypeptides; nucleotide sequences; mutants; herbicide resistance; atrazine; diuron; bromacil; binding site; amino acid sequences; chlorophyll, fluorescence; electron transfer

DNAL Call No.: QK725.P532

41

Characterization of a Chlamydomonas Transposon, Gulliver, Resembling those in Higher Plants

Ferris, P.J.

Source: GENETICS 122(2):363-377 (1989).

Descriptors: chlamydomonas reinhardtii; chromosome analysis; linkage maps; molecular genetics; cloning; deletions; chromosome maps; nucleotide sequence

DNAL Call No.: 442.8 G28

Abstract:

While pursuing a chromosomal walk through the mt(+) locus of linkage group VI of Chlamydomonas reinhardtii, I encountered a 12-kb sequence that was found to be present in approximately 12 copies in the nuclear genome. Comparison of various C. reinhardtii laboratory strains provided evidence that the sequence was mobile and therefore a transposon. One of two separate natural isolates interfertile with C. reinhardtii, C. smithii (CC-1373), contained the transposon, but at completely different locations in its nuclear genome than C. reinhardtii; and a second, CC-1952 (s1-C5) lacked the transposon altogether. Genetic analysis indicated that the transposon was found at dispersed sites throughout the genome, but had a conserved structure at each location. Sequence homology between the termini was limited to an imperfect 15-bp inverted repeat. An 8-bp target site duplication was created by insertion; transposon sequences were completely removed upon excision leaving behind both copies of the target site duplication, with minor base changes. The transposon contained an internal region of unique repetitive sequence responsible for restriction fragment length heterogeneity among the various copies of the transposon. In several cases it was possible to identify which of the dozen transposons in a given strain served as the donor when a transposition event occurred. The transposon often moved into a site genetically linked to the donor, and transposition appeared to be nonreplicative. Thus the mechanism of transposition and excision of the transposon, which I have named Gulliver, resembles that of certain higher plant transposons, like the Ac transposon of maize.

42

The 5'-Flanking Region of the Gene Encoding the Large Subunit of Ribulose-1,5-bisphoshate Carboxylase/oxygenase is Crucial for Growth of the Cyanobacterium Synechococcus sp. Strain PCC 7942 at the Level of CO2 in Air

Friedberg, D.; Kaplan, A.; Ariel, R.; Kessel, M.; Seijffers, J.

Source: JOURNAL OF BACTRIOLOGY 171(11):6069-6076 (1989).

Descriptors: synechococcus; strains; growth; carbon dioxide; ribulose-bisphosphate carboxylase; mutants; nucleotide sequence

DNAL Call No.: 448.3 J82

Abstract:

Transformation of the high-CO2-requiring mutants (hcr) 0221 and E1 derived from the cyanobacterium Synechococcus sp. strain PCC 7942 by a wild-type DNA library restored their ability to grow at the level of CO2 in air. A plasmid (pE12) containing a 10-kilobase DNA insert was rescued from a 0221 heterogenote and proved to transform both 0221 and E1 to the wild-type phenotype. The capacity of the pE12 subclones to confer the wild-type phenotype to 0221 transformants enabled the mapping of the mutation in 0221 (designated hcr0221) within a 232-base-pair PstI-BstXI DNA restriction fragment. Sequence analysis revealed two open reading frames (ORFs) at positions -1745 to -1262 (ORFI) and -1218 to -393 (ORFII) upstream of the rbcL gene. A 3-kilobase PstI fragment of 0221 was cloned, and hcr0221 was found to be a point mutation within the PstI-BstXI region -1309 nucleotides upstream of the rbcL gene. The significance of this flanking region for adaptation to air levels of CO2 was further demonstrated by the generation of new hcr mutants following insertional inactivation of wild-type DNA in the BstXI site. Electron microscopy revealed aberrant carboxysome structures in growing cells of the hcr mutants, a defect that was possibly related to the mutation, since transformation with pE12 derivatives restored the carboxysome structure to normal.

43

Red Algal Plasmids

Goff, L.J. and A.W. Coleman

Source: CURRENT GENETICS 18(6):557-565 (1990).

Descriptors: rhodophyta; plasmids; nucleotide sequences; amino acid sequences; dna; biotechnology

DNAL Call No.: QH426.C8

44

Transgenic Expression of Aminoglycoside Adenine Transferase in the Chloroplast a Selectable Marker for Site-Directed Transformation of Chlamydomonas

Goldschmidt-Clermont, M.

Source: NUCLEIC ACIDS RESEARCH 19(15):4083-4090 (1991).

Descriptors: chlamydomonas reinhardtii; bacterial aada gene transformation; vectors; transcription; translation

DNAL Call No.: AD341.A2N8

Abstract:

Expression vectors for Chlamydomonas reinhardtii chloroplast transformation have been constructed with transcription and translation signals from chloroplast genes. The bacterial aadA sequence coding for aminoglycoside 3" adenyl transferase, was inserted in these vectors and introduced into the C. reinhardtii chloroplast by particle gun transformation. The stable transgenic expression of this foreign protein in the chloroplast confers spectinomycin and streptomycin resistance to the transformed cells. This new marker can be used as a reporter of gene expression, and as a portable selectable cassette for chloroplast reverse genetics. Targetted gene disruption mutants of loci required for

photosynthisis, tscA and psaC, were thus obtained. A gene disruption of an unidentified open reading frame, ORF472, remained heteroplasmic, suggesting that it has a vital function.

45

Trans-splicing Mutants of Chlamydomonas reinhardtii
Goldschmidt-Clermont, M.; Girard-Bascou, J.; Choquet, Y.; Rochaix, J.D.
Source: MGG: MOLECULAR AND GENERAL GENETICS 223(3):417-425 (1990).
Descriptors: chlamydomonas reinhardtii; chloroplasts; genomes; plant proteins; photosystem i; genes; introns; exons; loci; messenger rna; alternative splicing; mutants; deletions; restriction mapping; northern blotting; segregation; recombination; complementation
DNA Call No.: 442.8 Z34

46

Gabaculine-Resistant Glutamate 1-Semialdehyde Aminotransferase of Synechococcus. Deletion of a Tripeptide Close to the NH2 Terminus and Internal Amino Acid Substitution.
Grimm, B.; Smith, A.J.; Kannangara, C.G.; Smith, M.
Source: THE JOURNAL OF BIOLOGICAL CHEMISTRY 266(19):12495-12501 (1991).
Descriptors: synechococcus; aminotransferases; glutamic acid; aminolevulinic acid; mutants; genes; cloning; nucleotide sequences; aminolevulinic acid; mutants; genes; cloning; nucleotide sequences
DNAL Call No.: 381 J824
Abstract:

Glutamate 1-semialdehyde aminotransferase (GSA-AT) is the last enzyme in the C5 pathway converting glutamate into the tetrapyrrole precursor delta-aminolevulinate in plants, algae, and several bacteria. Sequence analysis of the genes encoding GSA-AT in barley, Synechococcus, and Escherichia coli revealed 50-70% similarity in the primary structures of the proteins. The enzyme is inhibited rapidly by gabaculine when added in approximately stoichiometric amounts with the enzyme. A gabaculine-tolerant Synechococcus strain, GR6, was found to produce a GSA-AT less sensitive to the inhibitor. Accordingly, the mutant gene was isolated and sequenced. In comparison with the wild-type gene it contains a deletion of nine nucleotides (position 12-20) and a guanine to adenine substitution (position 743). This resulted in the loss of the amino acids serine, proline, and phenylalanine (position 5-7) close to the NH2 terminus of the enzyme and an exchange of Met-248 for isoleucine in the middle of the polypeptide chain. Wild-type and mutant GSA-AT were expressed in E.coli and purified close to homogeneity. Although the specific activity of the mutant GSA-AT was only one-fifth of the wild type, it displayed a 100-fold increased resistance to gabaculine. Peaks in the absorption spectrum of the purified recombinant GSA-ATs at 335 and 417 nm are typical of a transaminase containing a B6 cofactor. Incubation with substrate and with inhibitor induced spectral changes characteristic of other gabaculine-sensitive, B6-requiring enzymes.

47

Escherichia-coli and Anacystis-nidulans Plasmid Shuttle Vectors Containing the P-L Promoter from Bacteriophage Lambda
Gruber, M.Y.; Glick, B.R.; Thompson, J.E.
Source: CURRENT MICROBIOLOGY 22(1):15-20 (1991).
Descriptors: temperature-sensitive; CI857 repressor gene; genetic engineering; temperature regulated; foreign gene expression
DNAL Call No.: QR1.C78
Abstract:

Escherichia coli-Anacystis nidulans shuttle vectors pHIEX14, pSMG1, and pANH1,

containing the leftward promoter, PL, of bacteriophage lambda and the gene for the temperature-sensitive repressor, cI857, were constructed and used to transform A. nidulans. The transformation efficiencies and restriction endonuclease maps of these plasmids are reported. The use of these shuttle vectors should allow temperature regulation of foreign gene expression in A. nidulans.

48

Self-splicing of the Chlamydomonas Chloroplast psbA Introns

Herrin, D.L.; Bao, Y.; Thompson, A.J.; Chen, Y.F.

Source: THE PLANT CELL 3(10):1095-1107 (1991).

Descriptors: chlamydomonas; introns; alternative splicing; transcription; photosystem ii; plant proteins; genes; chloroplast genetics; nucleotide sequences; chloroplasts; genomes

DNAL Call No.: QK725.P532

Abstract:

We used alpha-(32)P-GTP labeling of total RNA preparations to identify self-splicing group I introns in Chlamydomonas. Several RNAs become labeled with alpha-(32)P-GTP, a subset of which is not seen with RNA from a mutant that lacks both copies of the psbA gene. Hybridization of the GTP-labeled RNAs to chloroplast DNA indicates that they originate from the psbA and rrn 23s genes, respectively, the only genes known to contain group I introns in this organism. Introns 1, 2, and 3 of psbA (with flanking exon sequences) were subcloned and transcribed in vitro. The synthetic RNAs were found to self-splice; splicing required Mg2+, GTP, and elevated temperature. In addition, the accuracy of self-splicing was confirmed for introns 1 and 2, and intermediates in the splicing reactions were detected. These results, together with our recent data on the 23S intron, indicate that the ability to self-splice is a general feature of Chlamydomonas group I introns. These findings have significant implications for the mechanism of group I intron splicing and evolution in Chlamydomonas and other chloroplast genomes.

49

RNA Splicing in Chlamydomonas Chloroplasts. Self-splicing of 23 S preRNA

Herrin, D.L.; Chen, Y.F.; Schmidt, G.W.

Source: THE JOURNAL OF BIOLOGICAL CHEMISTRY 265(34):21134-21140 (1990).

Descriptors: chlamydomonas reinhardtii; chloroplast genetics; rna; precursors; introns; gene mapping; transcription

DNAL Call No.: 381 J824

50

Cloning and Expression of the Chloroplast-Encoded rbcL and rbcS Genes from the Marine Diatom Cylindrotheca sp. strain N1

Hwang, S.R. and Tabita, F.R.

Source: PLANT MOLECULAR BIOLOGY: AN INTERNATIONAL JOURNAL ON MOLECULAR BIOLOGY, BIOCHEMISTRY AND GENETIC ENGINEERING 13(1):69-79 (1989).

Descriptors: bacillariophyta; escherichia coli; anacystis nidulans; multiple genes; ribulose-bisphosphate carboxylase; genomes; chloroplast genetics; gene expression; cloning; gene mapping; restriction mapping; recombinant dna; gene splicing; enzyme activity

DNAL Call No.: QK710.P62

Abstract:

Both the rbcL and rbcS genes, encoding the large and small subunits, respectively, of ribulose 1,5-bisphosphate carboxylase/oxygenase, have been found to be encoded by chloroplast DNA in the marine diatom Cylindrotheca sp. N1. The rbcS gene in this diatom was found to be adjacent to the rbcL gene by a combination of: (i) Southern-

blotting analyses, using heterologous probes; (ii) examination of recombinant proteins synthesized in Escherichia coli, directed by cloned rbcL/rbcS genes; and (iii) synthesis of enzymatically active heterologous Rubisco protein in vivo by recombinant DNA procedures using large subunits of Anacystis nidulans and small subunits of Cylindrotheca sp. N1. It appears that two copies of rbcL and rbcS genes are encoded by the chloroplast DNA of this diatom.

51

Cloning of the psbK Gene from Synechocystis sp. PCC 6803 and Characterization of Photosystem II in Mutants Lacking PSII-K

Ikeuchi, M.; Eggers, B.; Shen, G.; Webber, A.; Yu, J.; Hirano, A.; Inoue, Y.; Vermaas, W.

Source: THE JOURNAL OF BIOLOGICAL CHEMISTRY 266(17):11111-11115 (1991).

Descriptors: cyanobacteria; mutants; photosystem ii; genetic engineering; plant proteins; cloning; nucleotide sequences

DNAL Call No.: 381 J824

Abstract:

We cloned and sequenced the psbK gene, coding for a small photosystem II component (PSII-K), from the transformable cyanobacterium, Synechocystis sp. PCC 6803, and determined the N-terminal sequence of mature PSII-K. The psbK gene product is processed by cleaving off eight amino acid residues from the N terminus. A mutant lacking psbK was constructed; this mutant grew photoautotrophically, but its growth rate was reduced. The number of photosystem II reaction centers on a chlorophyll basis was decreased by less than a factor of 2 in the psbK-deletion mutant. In Synechocystis sp. PCC 6803, the psbK gene is transcribed as a single gene and is not part of an operon. Single-site mutations were introduced into psbK leading to early termination or deletion of the presequence. The phenotype of these mutants strongly resembles that of the psbK deletion mutant, indicating that indeed the change in phenotype in the deletion mutant is directly correlated with PSII-K. PSII-K is not essential for photosystem II assembly or activity but is needed for optimal photosystem II function.

52

Splice Site Selection and Role of the Lariat in a Group II Intron

Jacquier, A. and Jacquesson-Breuleux, N.

Source: JOURNAL OF MOLECULAR BIOLOGY 219(3):415-428 (1991).

Descriptors: fungi; algae; plants; rna; molecular conformation; introns; catabolism; hydrolysis; mutants

DNAL Call No.: 442.8 J8224

Abstract:

The structural elements involved in 5' and 3' splice site (SS) selection in a group II intron were analyzed. While 5' SS selection appears to be defined by only one element, the EBS1-IBS1 pairing, four distinct structural components contribute to 3' SS selection, one of which being analogous to the "internal guide sequence" described for group I introns. Moreover, some of the mutants analyzed during this study induce efficient 5' SS hydrolysis and suggest how 5' SS transesterification is selected against hydrolysis. Finally, the lariat structure was found to accelerate both steps of splicing, suggesting that is "locks" the ribozyme in an active configuration.

53

Transient Expression of Firefly Luciferase in Protoplasts of the Green Alga Chlorella-ellipsoidea

Jarvis, E.E. and Brown, L.M.

Source: CURRENT GENETICS 19(4):317-322 (1991).

Descriptors: dna; transformation; genetic engineering

DNAL Call No.: QH426.C8
Abstract:

We report here on the development of a transient expression system for Chlorella ellipsoidea using a heterologous gene, firefly luciferase. Cells of this unicellular green alga were converted to protoplasts and treated with plasmid pD0432, which bears luciferase under the control of the CaMV 35s promoter. This treatment resulted in detectable luciferase activity in cell extracts. Expression required Cellulysin treatment, active cell metabolism, and the addition of carrier DNA and polyethylene glycol. Linearization of the luciferase plasmid did not significantly alter the activity. A time course of expression showed that luciferase is made rapidly, within about 7 h after addition of DNA, but that the activity disappears over the course of a few days. These experiments represent an important first step in the development of a Chlorella transformation system.

54

Molecular Studies of Linkage Group XIX of Chlamydomonas reinhardtii: Evidence Against a Basal Body Location

Johnson, D.E. and Dutcher, S.K.

Source: THE JOURNAL OF CELL BIOLOGY 113(2):339-346 (1991).

Descriptors: chlamydomonas reinhardtii; dna; linkage groups; restriction fragment length polymorphism; organelles; flagella; repetitive dna; transposable elements; dna hybridization

DNAL Call No.: 442.8 J828

Abstract:

Linkage group XIX (also known as the UNI linkage group) in the green alga, Chlamydomonas reinhardtii, exhibits a number of unusual properties that have lead to the suggestion that it represents a basal body-associated chromosome. To begin a molecular analysis of this linkage group, we have identified DNA sequences from it and used them to determine the copy number of linkage group XIX within the cell. We find that linkage group XIX is present in the same copy number per cell as nuclear linkage groups in both haploid and diploid strains. We also find that the copy number of linkage group XIX is unchanged in mutants lacking basal bodies. We conclude that there is no convincing evidence that linkage group XIX localizes to the basal bodies of Chlamydomonas reinhardtii cells.

55

Expression of Salmon Growth Hormone in the Cyanobacterium Agmenellum-quadruplicatum

Kawata, Y.; Yamano, N.; Kojima, H.; Itoh, S.

Source: BIOTECHNOLOGY LETTERS 13(12):851-856 (1991).

Descriptors: escherichia-coli; bacteria; microorganism; fish genetics; gene transfer; TRP promoter; total cell protein; aquaculture; biotechnology industry

DNAL Call No.: QR53 B56

Abstract:

The salmon growth hormone gene was introduced into the cyanobacterium Agmenellum quadruplicatum PR-6 by plasmid transformation. The gene expressed the hormone under the trp promoter of Escherichia coli. The amount was estimated to be approximately 0.1% of the total cell protein.

56

Engineering the Chloroplast Genome: Techniques and Capabilities for Chloroplast Transformation in Chlamydomonas reinhardtii

Kindle, K.L.; Richards, K.L.; Stern, D.B.

Source: PROCEEDINIGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE

UNITED STATES OF AMERICA 88(5):1721-1725 (1990).
Descriptors: chlamydomonas reinhardtii; chloroplast genetics; genetic engineering; genetic transformation; genomes; methodology
DNAL Call No.: 500 N21P
Abstract:

Chloroplast transformation of Chlamydomonas reinhardtii has been accomplished by agitating cell wall-deficient cells in the presence of glass beads and DNA. By using the atpB gene as the selected marker and cells grown in 0.5 mM 5-fluorodeoxyuridine, we have recovered up to 50 transformants per microgram of DNA. This method is easy and does not require specialized equipment, although it is not as efficient as the tungsten particle bombardment method [Boynton, J.E., Gillham, N.W., Harris, E.H., Hosler, J.P., Johnson, A.M., Jones, A.R., Randolph-Anderson, B.L., Robertson, D., Klein, T.M., Shark, K.B. and Sanford, J.C. (1988) Science 240, 1534-1537]. By using particle bombardment, we have developed a cotransformation approach in which spectinomycin-resistant 16S rRNA-encoding DNA is the selected marker, and we have demonstrated that cotransformation of an unselected marker on an independent replicon is very efficient. We have used this strategy (i) to recover transformants with partially deleted atpB genes that could not otherwise have been selected since they did not restore photosynthetic capability to a recipient carrying a more extensive atpB deletion and (ii) to generate specific deletion mutations in a wild-type recipient. This methodology should allow the introduction of any desired change into the chloroplast genome, even in the absence of phenotypic selection, and thus a detailed functional analysis of any chloroplast DNA sequence should be possible.

57
High-Frequency Nuclear Transformation of Chlamydomonas-reinhardtii
Kindle, K.L.
Source: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA 87(3):1228-1232 (1990).
Descriptors: photosynthesis; chloroplast; biogenesis; dna; transfection; genetic engineering
DNAL Call No.: 500 N21P
Abstract:

By using a method in which cell-wall-deficient Chlamydomonas reinhardtii cells were agitated in the presence of DNA, glass beads, and polyethylene glycol, nuclear transformation rates of .apprxeq. 103 transformations per .mu.g of plasmid DNA were achieved. The nitrate reductase gene from wild-type Chlamydomonas was used to complement a mutation in the corresponding gene of a strain containing nit1-305. Transformants were selected by growth with nitrate as sole source of nitrogen. The transforming DNA integrated into the genome at a low-copy number in nit+ transformants. When cells carrying nit1-305 were agitated in the presence of two plasmids, one with the gene for nitrate reductase and the second with an unselected gene, the unselected gene was present in 10-50% of nit+ transformants. This high frequency of cotransformation will allow any cloned gene to be introduced into Chlamydomonas. Moreover, the overall efficiency of transformation should be high enough to permit isolation of genes from genomic libraries by complementation of stable nuclear mutants. The availability of efficient nuclear and chloroplast transformation in Chlamydomonas provides specific advantages for the study of chloroplast biogenesis, photosynthesis, and nuclear-chloroplast genome interactions.

58
The Cyanelle str Operon from Cyanophora paradoxa: Sequence Analysis and Phylogenetic Implications

Kraus, M.; Gotz, M.; Loffelhardt, W.
Source: PLANT MOLECULAR BIOLOGY: AN INTERNATIONAL JOURNAL ON MOLECULAR BIOLOGY, BIOCHEMISTRY AND GENETIC ENGINEERING 15(4):561-573 (1990).
Descriptors: algae; genes; ribosomes; plant proteins; cloning; nucleotide sequences; organelles; phylogeny; amino acid sequences; transcription; messenger rna
DNAL Call No.: QK710.P62
Abstract:

The str operon containing the genes for the ribosomal proteins S12 (rps12) and S7 (rps7) and for the elongation factors G (fus) and Tu (tufA) has been characterized for some cyanobacteria and chloroplasts from algae and higher plants. In the case of plastids a stepwise reduction by one and two genes, respectively, has been observed due to gene transfer to the nuclear genome. The nucleotide sequence of the str operon on the cyanelle genome from Cyanophora paradoxa was determined as a first example for a chlorophyll b-less plastid. It comprises rps12, rps7 and tufA which are closely linked and not interrupted by introns. Transcript analysis revealed cotranscription of the two ribosomal protein genes whereas tufA gave rise to a monocistronic mRNA. Phylogenetic studies using these three different traits allowed an assessment of the position of Cyanophora paradoxa among oxygenic photoautotrophs.

59
Conjugative Transfer and Autonomous Replication of a Promiscuous IncQ Plasmid in the Cyanobacterium Synechocystis PCC 6803
Kreps, S.; Ferino, F.; Mosrin, C.; Gerits, J.; Mergeay, M.; Thuriaux, P.
Source: MGG: MOLECULAR AND GENERAL GENETICS 221(1):129-133 (1990).
Descriptors: cyanobacteria; escherichia coli; saccharomyces cerevisiae; plasmids; genetic transformation; cloning; photosynthesis
DNAL Call No.: 442.8 Z34

60
Developmental Rearrangement of Cyanobacterial nif Genes: Nucleotide Sequence, Open Reading Frames, and Cytochrome P-450 Homology of the Anabaena sp. Strain PCC 7120 nifD Element
Lammers, P.J.; McLaughlin, S.; Papin, S.; Trujillo-Provencio, C.; Ryncarz, A.J. II
Source: JOURNAL OF BACTERIOLOGY 172(12):6981-6990 (1990).
Descriptors: anabaena; strains; genes; nucleotide sequences; amino acid sequences; cytochrome p-450
DNAL Call No.: 448.3 J82
Abstract:

An 11-kbp DNA element of unknown function interrupts the nifD gene in vegetative cells of Anabaena sp. strain PCC 7120. In developing heterocysts the nifD element excises from the chromosome via site-specific recombination between short repeat sequences that flank the element. The nucleotide sequence of the nifH-proximal half of the element was determined to elucidate the genetic potential of the element. Four open reading frames with the same relative orientation as the nifD element-encoded xisA gene were identified in the sequenced region. Each of the open reading frames was preceded by a reasonable ribosome-binding site and had biased codon utilization preferences consistent with low levels of expression. Open reading frame 3 was highly homologous with three cytochrome P-450 omega-hydroxylaseproteins and showed regional homology to functionally significant domains common to the cytochrome P-450 superfamily. The sequence encoding open reading frame 2 was the most highly conserved portion of the sequenced region based on heterologous hybridization experiments with three genera of heterocystous cyanobacteria.

61

Genomic Structure of Chlamydomonas caltractin. Evidence for Intron Insertion Suggests a Probable Genealogy for the EF-Hand Superfamily of Proteins

Lee, V.D.; Stapleton, M.; Huang, B.

Source: JOURNAL OF MOLECULAR BIOLOGY 221(1):175-191 (1991).

Descriptors: chlamydomonas reinhardtii; genes; calcium binding proteins; genome analysis; introns; exons; nucleotide sequences; amino acid sequences; evolution; ancestry; structure; molecular conformation

DNAL Call No.: 442.8 J8224

Abstract:

A clone containing the gene locus for Chlamydomonas caltractin, a 20,000 Mr calcium-binding protein that is a member of the EF-hand superfamily of calcium-modulated proteins, was isolated and the structural organization of the gene was determined. The intron-exon organization was resolved by direct comparison of the genomic sequence with a caltractin cDNA. The promoter region does not contain the typical TATA or CCAAT boxes, but the sequences at the splice junctions are similar to those of other eukaryotes. The positions of the six introns in the caltractin gene do not typically define unit structures, nor do they coincide with those in genes for other members of the EF-hand superfamily. An analysis of exon sequences at the splice junctions in the genes of this multigene family was undertaken; evidence was obtained that supports the hypothesis that introns arose at protosplice sites. A probable evolutionary history for the EF-hand superfamily based on intron insertion is offered.

62

Recombination of Chlamydomonas Chloroplast DNA Occurs more Frequently in the Large Inverted Repeat Sequence than in the Single-copy Regions

Lemieux, B.; Turmel, M.; Lemieux, C.

Source: THEORETICAL AND APPLIED GENETICS 79(1):17-27 (1990).

Descriptors: chlamydomonas; hybrids; chloroplast genetics; dna; recombination; genetic code; nucleotide sequence; inheritance; genetic polymorphism; gene mapping

DNAL Call No.: 442.8 Z8

63

Homologues of the Green Algal gidA Gene and the Liverwort frxC Gene are Present on the Chloroplast Genomes of Conifers

Lidholm, J. and Gustafsson, P.

Source: PLANT MOLECULAR BIOLOGY: AN INTERNATIONAL JOURNAL ON MOLECULAR BIOLOGY, BIOCHEMISTRY AND GENETIC ENGINEERING 17(4):787-798.

Descriptors: pinus contorta; picea abies; chlamydomonas reinhardtii; marchantia polymorpha; genomes; dna; chloroplasts; nucleotide sequences; dna hybridization; southern blotting; genes; transfer rna; asparagine; amino acid sequences; chlorophyll; biosynthesis

DNA Call No.: QK710.P62

Abstract:

Strong hybridization signals were obtained from total DNA of two conifers, lodgepole pine (Pinus contorta) and Norway spruce (Picea abies), in a Southern blot analysis using a probe derived from the chloroplast gidA gene of the green alga Chlamydomonas reinhardtii. The pine fragments detected by the probe were found to originate from the chloroplast genome and, as judged by the signal intensity, this was also true for the spruce fragments. Sequence analysis of the hybridizing pine chloroplast DNA region revealed an open reading frame potentially encoding a 459 amino acid polypeptide, highly homologous to that deduced from the algal gene and to ORF465 of liverwort chloroplast DNA. Upstream of the gidA sequence, we found a trnN(GUU) gene and an open reading frame

of 291 codons which was 78% identical to the frxC gene of liverwort. Since ORF465 is located immediately downstream of trnN and frxC in liverwort, the genetic organization of this region is very similar in the two plants. In contrast, neither the gidA nor the frxC gene is present in the chloroplast DNA of tobacco or rice. It was recently reported that deletions in the gidA region of the chloroplast genome of Chlamydomonas reinhardtii abolish the light-independent pathway of chlorophyll synthesis which exists in many algae and lower plants. The presence of the gidA gene on the chloroplast genomes of conifers may therefore be of significance with respect to the ability of these plants to synthesize chlorophyll in the dark.

64

Structural Features of the Plastid Ribosomal RNA Operons of Two Red Algae: Antithamnion sp. and Cyanidium caldarium

Maid, U. and Zetsche, K.

Source: PLANT MOLECULAR BIOLOGY: AN INTERNATIONAL JOURNAL ON FUNDAMENTAL RESEARCH AND GENETIC ENGINEERING 16(4):537-546 (1991).

Descriptors: rhodophyta; ribosomal rna; ribosomal dna; nucleotide sequences; chloroplasts; evolution; chemotaxonomy; genes; transfer rna; isoleucine; alanine

DNAL Call No.: QK710.P62

Abstract:

The nucleotide sequences of the plastid 16S rDNA of the multicellular red alga Antithamnion sp. and the 16S rDNA/23S rDNA intergenic spacers of the plastid DNAs of the unicellular red alga Cyanidium caldarium and of Antithamnion sp. were determined. Sequence comparisons support the idea of a polyphyletic origin of the red algal and the higher-plant chloroplasts. Both spacer regions include the unsplit tRNA (Ile) (GAU) and TRNA (Ala) (UGC) genes and so the plastids of both algae form a homogeneous group with those of chromophytic algae and Cyanophora paradoxa characterized by 'small-sized' rDNA spacers in contrast to green algae and higher plants. Nevertheless, remarkable sequence differences within the rRNA and the tRNA genes give the plastids of Cyanidium caldarium a rather isolated position.

65

Characterization of Cryptic Plasmids from Marine Cyanobacteria and Construction of a Hybrid Plasmid Potentially Capable of Transformation of Marine Cyanobacterium Synechococcus-sp and its Transformation

Matsunaga, T.; Takeyama, H.; Nakamura, N.

Source: APPLIED BIOCHEMISTRY AND BIOTECHNOLOGY 24-25(spring-summer):151-160 (1990).

Descriptors: anacystis-nidulans; escherichia-coli; electroporation; genetic engineering; biotechnology

DNAL Call No.: QD415.A1J62

66

Dynamic Interplay between two Copper-Titrating Components in the Transcriptional Regulation of cyt c6

Merchant, S.; Hill, K.; Howe, G.

Source: THE EMBO JOURNAL-EUROPEAN MOLECULAR BIOLOGY ORGANIZATION 10(6):1383-1389 (1991).

Descriptors: chlamydomonas reinhardtii; transcription; gene expression; genetic regulation; cytochrome c; genes; copper; binding proteins; messenger rna; plastocyanins

DNAL Call No.: QH506.E46

Abstract:

The algal plastidic cytochrome c (cyt c6) is a biochemical equivalent of the copper-containing protein plastocyanin in photosynthetic electron transfer. But generally, cyt c6 accumulates and functions only under conditions (e.g. Cu-deficiency) where holoplastocyanin cannot be synthesized. In studying the regulation of Chlamydomonas reinhardtii cyt c6 expression by Cu we have determined that repression of cyt c6 accumulation occurs at the transcriptional level, and specifically in response to Cu as the metal ion regulator. Complete and sustained repression of cyt c6 transcription requires approximately 9 X 10(6) Cu ions in the medium/cell. Based on the estimated plastocyanin content of algal cells (8 X 10(6) molecules/cell) and the observation that lower ratios of Cu per cell result in only transient repression of cyt c6 transcription, we propose that Cu-dependent transcriptional repression of the gene encoding cyt c6 requires a Cu-binding factor which is titrated by Cu only after the alternate electron transfer catalyst, plastocyanin, has accumulated to the stoichiometry required for photosynthesis. The precise and highly metal-specific, autoregulatory control of cyt c6 levels--directly by Cu, and indirectly by holoplastocyanin--is in keeping with the functional role of cyt c6 as an alternate, although perhaps less preferred, electron transfer catalyst.

67

Targetted Disruption of Chloroplast Genes in Chlamydomonas reinhardtii

Newman, S.M.; Gillham, N.W.; Harris, E.H.; Johnson, A.M.; Boynton, J.E.

Source: MGG: MOLECULAR AND GENERAL GENETICS 230(1/2):65-74 (1991).

Descriptors: chlamydomonas reinhardtii; chloroplast genetics; genetic transformation; plasmids; gene transfer; mutations; genes; direct dna uptake; targeted mutagenesis

DNAL Call No.: 442.8 Z34

Abstract:

We have developed an efficient procedure for the disruption of Chlamydomonas chloroplast genes. Wild-type C. reinhardtii cells were bombarded with microprojectiles coated with a mixture of two plasmids, one encoding selectable, antibiotic-resistance mutations in the 16S ribosomal RNA gene and the other containing either the atpB or rbcL photosynthetic gene inactivated by an insertion of 0.48 kb of yeast DNA in the coding sequence. Antibiotic-resistant transformants were selected under conditions permissive for growth of non-photosynthetic mutants. Approximately half of these transformants were initially heteroplasmic for copies of the disrupted atpB or rbcL genes integrated into the recipient chloroplast genome but still retained photosynthetic competence. A small fraction of the transformants (1.1% for atpB; 4.3% for rbcL) were nonphotosynthetic and homoplasmic for the disrupted gene at the time they were isolated. Single cell cloning of the initially heteroplasmic transformants also yielded nonphotosynthetic segregants that were homoplasmic for the disrupted gene. Polypeptide products of the disrupted atpB and rbcL genes could not be detected using immunoblotting techniques. We believe that any nonessential Chlamydomonas chloroplast gene, such as those involved in photosynthesis, should be amenable to gene disruption by cotransformation. The method should prove useful for the introduction of site-specific mutations into chloroplast genes and flanking regulatory sequences with a view to elucidating their function.

68

Transformation of Chloroplast Ribosomal RNA Genes in Chlamydomonas: Molecular and Genetic Characterization of Integration Events

Newman, S.M.; Boynton, J.E.; Gillham, N.W.; Randolph-Anderson, B.L.; Johnson, A.M.; Harris, E.H.

Source: GENETICS 126(4):875-888 (1990).

Descriptors: chlamydomonas reinhardtii; chlamydomonas; ribosomal dna; ribosomal rna; genes;

chloroplasts; genomes; genetic transformation; induced mutations; direct dna uptake; phenotypes; antibiotics; drug resistance; gene mapping; restriction fragment length polymorphism
DNAL Call No.: 442.8 G28
Abstract:

Transformation of chloroplast ribosomal RNA (rRNA) genes in Chlamydomonas has been achieved by the biolistic process using cloned chloroplast DNA fragments carrying mutations that confer antibiotic resistance. The sites of exchange employed during the integration of the donor DNA into the recipient genome have been localized using a combination of antibiotic resistance mutations in the 16S and 23S rRNA genes and restriction fragment length polymorphisms that flank these genes. Complete or nearly complete replacement of a region of the chloroplast genome in the recipient cell by the corresponding sequence from the donor plasmid was the most common integration event. Exchange events between the homologous donor and recipient sequences occurred preferentially near the vector:insert junctions. Insertion of the donor rRNA genes and flanking sequences into one inverted repeat of the recipient genome was followed by intramolecular copy correction so that both copies of the inverted repeat acquired identical sequences. Increased frequencies of rRNA gene transformants were achieved by reducing the copy number of the chloroplast genome in the recipient cells and by decreasing the heterology between donor and recipient DNA sequences flanking the selectable markers. In addition to producing bona fide chloroplast rRNA transformants, the biolistic process induced mutants resistant to low levels of streptomycin, typical of nuclear mutations in Chlamydomonas.

69
A Gene Homologous to the Subunit-2 Gene of NADH Dehydrogenase is Essential to Inorganic Carbon Transport of Synechocystis PCC6803
Ogawa, T.
Source: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA 88(10):4275-4279 (1991).
Descriptors: cyanobacteria; amino acid sequences; chloroplasts; mitochondria; mutants; translocation; carbon; carbon dioxide; genetic transformation; nadh dehydrogenase; nucleotide sequences; respiration; wild strains
DNAL Call No.: 500 N21P
Abstract:

A clone that transforms the RKa mutant of Synechocystis PCC6803 defective in inorganic carbon (Ci) transport to the wild-type phenotype was isolated from a cyanobacterial genomic library. The clone contained an 11.8-kilobase-pair DNA insert. Sequencing of the insert DNA in the region of the mutation in Rka revealed an open reading frame (designated as ndhB), which showed extensive amino acid sequence homology to the subunit-2 genes of NADH dehydrogenase (EC 1.6.99.3) (ndhB) of chloroplasts and mitochondria. The homology was much stronger with the chloroplast genes. Sequence analysis of the ndhB gene of RKa mutant revealed a G leads to A substitution that results in a Gly lead to Asp substitution in the deduced amino acid. A defined mutant (M55), constructed by inactivating the ndhB gene in wild-type Synechocystis, required high CO2 conditions for growth and was unable to transport CO2 and HCO3- into the intracellular Ci pool. The results indicate that the ndhb gene is required for Ci transport. Dark respiration was also depressed by the inactivation of the ndhB gene. A possible role of the ndhB gene product in the energization of Ci transport is discussed.

70
Recombination: Recombination of Mobile Genetic Elements from Plants and Cyanobacteria
Osiewacz, H.D. and Heinen, U.

Source: PROGRESS IN BOTANY=FORTSCHRITT DER BOTANIK 50:174-197 (1989).
Descriptors: plants; recombination; gene expression; dna
DNAL Call No.: 450 F772

71
Detection of Gene Expression in Genetically Engineered Microorganisms and Natural Phytoplankton Populations in the Marine Environment by Messenger RNA Analysis
Pichard, S.L. and Paul, J.H.
Source: APPLIED AND ENVIRONMENTAL MICROBIOLOGY 57(6):1721-1727 (1991).
Descriptors: vibrio synechococcus; plasmid-coded; neomycin; phosphotransferase NPTII gene; ribulose bisphosphate carboxylase-oxygenase; large subunit RBCL gene; diurnal expression pattern; dry tortugas florida usa
DNAL Call No.: 448.3 AP5
Abstract:

A simple method that combines guanidinium isothiocyanate RNA extraction and probing with antisense and sense RNA probes is described for analysis of microbial gene expression in planktonic populations. Probing of RNA sample extracts with sense-strand RNA probes was used as a control for nonspecific hybridization or contamination of mRNA with target DNA. This method enabled detection of expression of a plasmid-encoded neomycin phosphotransferase gene (nptII) in as few as 105 Vibrio cells per ml in 100 ml of seawater. We have used this method to detect expression of the ribulose-1,5-bisphosphate carboxylase large-subunit gene (rbcL) in Synechococcus cultures and natural phytoplankton populations in the Dry Tortugas, Florida. During a 36-h diel study, rbcL expression of the indigenous phytoplankton was greatest in the day, least at night (1100, 0300, and 0100 h), and variable at dawn or dusk (0700 and 1900 h). These results are the first report of gene expression in natural populations by mRNA isolation and probing. This methodology should be useful for the study of gene expression in microorganisms released into the environment for agricultural or bioremediation purposes and indigenous populations containing highly conserved target gene sequences.

72
Intercistronic Group III Introns in Polycistronic Ribosomal Proteins of Chloroplasts
Stevenson, J.K.; Drager, R.G.; Copertino, D.W.; Christopher, D.A.; Jenkins, K.P.; Yepiz-Plascencia, G; Hallick, R.B.
Source: MGG: MOLECULAR AND GENERAL GENETICS 228(1/2):183-192 (1991).
Descriptors: euglena gracilis; introns; cistrons; genes; ribosomes; plant proteins; transcription; gene expression; messenger rna; nucleotide sequences; amino acid sequences; restriction mapping; chloroplasts; chloroplast genetics
DNAL Call No.: 442.8 Z34
Abstract:

A novel ribosomal protein operon in the Euglena gracilis chloroplast genome was characterized. It encodes the genes for ribosomal proteins S4 and S11 (rps4 and rps11). The coding region of the rps11 gene is interrupted by two introns of 107 and 100 bp. The introns belong to a distinct class known as group III introns. The major transcript from this operon was characterized as a fully spliced dicistronic rps4-rps11 mRNA by RNA blot analysis, primer extension sequencing, and cDNA cloning and sequencing. An additional 95 nucleotide (nt) group III intron was identified in the 123 nt rps4-rps11 intercistronic region. The identification of the intercistronic intron between the rps4 and rps11 genes was unexpected. Other RNA transcripts from regions of the genome that could potentially contain intercistronic introns were re-examined and two other intercistronic, group III introns were found. These are located in a large ribosomal protein operon between the genes for the ribosomal proteins L23 and L2, and between L14 and L5.

There are at least 50 group III introns in the E. gracilis chloroplast genome. All but 6 are found in genes encoding protein components of the transcriptional and translational apparatus. The distribution of group III introns and the unusual location of intercistronic group III introns may reflect some aspect of gene expression, or provide some insight into the mechanism of their splicing.

73

Directed Chloroplast Transformation in Chlamydomonas reinhardtii: Insertional Inactivation of the psaC Gene Encoding the Iron Sulfur Protein Destabilizes Photosystem I

Takahashi, Y.; Goldschmidt-Clermont, M.; Soen, S.Y.; Franzen, L.G.; Rochaix, J.D.

Source: THE EMBO JOURNAL-EUROPEAN MOLECULAR BIOLOGY ORGANIZATION 10(8):2033-2040 (1991).

Descriptors: chlamydomonas reinhardtii; chloroplasts; genes; photosystem i; nucleotide sequences; amino acid sequences; binding proteins

DNAL Call No.: QH506.E46

Abstract:

The chloroplast gene psaC encoding the iron sulfur protein of photosystem I (PSI) from the green alga Chlamydomonas reinhardtii has been cloned and characterized. The deduced amino acid sequence is highly, related to that of higher plants and cyanobacteria. Using a particle gun, wild type C. reinhardtiii cells have been transformed with a plasmid carrying the psaC gene disrupted by an aadA gene cassette designed to express spectinomycin/streptomycin resistance in the chloroplast. Transformants selected on plates containing acetate as a reduced carbon source and spectinomycin are unable to grow on minimal medium lacking acetate and are deficient in PSI activity. Southern blot analysis of total cell DNA of the transformants shows that the wild type psac gene has been replaced by the interrupted psaC gene through homologous recombination. While authentic transcripts of the psaC gene are no longer detected, aadA gives rise to a few transcripts in the transformants. Biochemical analysis indicates that neither PSI reaction center subunits nor the seven small subunits belonging to PSI accumulate stably in the thylakoid membranes of the transformants. Pulse-chase labeling of cell proteins shows that the PSI reaction center subunits are synthesized normally but turn over rapidly in the transformants. We conclude that the iron sulfur binding protein encoded by the psaC gene is an essential component, both for photochemical activity and for stable assembly of PSI. The present study suggests that any chloroplast gene encoding a component of the photosynthetic apparatus can be disrupted in C. reinhardtii using the strategy described.

74

Short Leader Sequences may be Transferred from Small RNAs to Pre-mature mRNAs by Trans-splicing in Euglena

Tessier, L.H.; Keller, M.; Chan, R.L.; Fournier, R.; Weil, J.H.; Imbault, P.

Source: THE EMBO JOURNAL- EUROPEAN MOLECULAR BIOLOGY ORGANIZATION 10(9):2621-2625 (1991).

Descriptors: euglena gracilis; messenger rna; rna; nucleotide sequences

DNAL Call No.: QH506.E46

Abstract:

Very closely related short sequences are present at the 5' end of cytoplasmic mRNAs in Euglena as evidenced by comparison of cDNA sequences and hybrid-arrested translation experiments. By cloning Euglena gracilis nuclear DNA and isolating the rbcS gene (encoding the small subunit of ribulose-1,5-bisphosphate carboxylase/oxygenase), we have shown that the short leader sequence does not flank the nuclear gene sequence. The leader sequences were found to constitute the 5' extremities of a family of small RNAs. Sequencing six members of this family revealed a striking similarity to vertebrate

UsnRNAs. We propose that a trans-splicing mechanism transfers the spliced leader (SL) sequence from these small RNAs (SL RNAs) to pre-mature mRNAs. Transfer of leader sequences to mRNAs by trans-splicing has been shown only in trypanosomes where cis-splicing is unknown, and in nematodes where not more than 10% of the mRNAs have leader sequences. Our results strongly suggest that Euglena is a unique organism in which both a widespread trans-splicing and a cis-splicing mechanism co-exist.

75

Photosynthetic Electron Transport Controls Nitrogen Assimilation in Cyanobacteria by Means of Posttranslational Modification of the glnB Gene Product

Tsinoremas, N.F.; Castets, A.M.; Harrison, M.A.; Allen, J.F.; Tandeau de Marsac, N.

Source: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA 88(11):4565-4569 (1991).

Descriptors: synechococcus; strains; amino acid sequences; ammonium; cloning; electron transfer; genetic code; molecular genetics; nitrogen metabolism; nucleotide sequences; photosynthesis; polypeptides; restriction mapping; transcription

DNAL Call No.: 500 N21P

Abstract:

A glnB gene is identified in the cyanobacterium Synechococcus sp. PCC 7942, and its gene product is found to be covalently modified as a result of imbalance in electron transfer in photosynthesis, where photosystem II is favored over photosystem I. The gene was cloned and sequenced and found to encode a polypeptide of 112 amino acid residues, whose sequence shows a high degree of similarity to the Escherichia coli regulatory protein, P(II). In E. coli, P(II) is involved in signal transduction in transcriptional and posttranslational regulation of nitrogen assimilation. Increase in ammonium ion concentration is shown to decrease covalent modification of the Synechococcus P(II) protein, as in enteric bacteria. We therefore propose that the photosynthetic electron transport chain may regulate the pathway of nitrogen assimilation in cyanobacteria by means of posttranslational, covalent modification of the glnB gene product. The existence of the glnB gene in different strains of cyanobacteria is demonstrated and its implications are discussed.

76

Six Group I Introns and Three Internal Transcribed Spacers in the Chloroplast Large Subunit Ribosomal RNA Genes of the Green Alga Chlamydomonas eugametos

Turmel, M.; Boulanger, J.; Schnare, M.N.; Gray, M.W.; Lemieux, C.

Source: JOURNAL OF MOLECULAR BIOLOGY 218(2):293-311 (1991).

Descriptors: chlamydomonas eugametos; chloroplast genetics; ribosomal rna; genes; nucleotide sequences; genome analysis; introns; transcription; nucleases

DNAL Call No.: 442.8 J8224

Abstract:

The chloroplast large subunit rRNA gene of Chlamydomonas eugametos and its 5' flanking region encoding tRNA(Ile) (GAU) and tRNA(Ala) (UGC) have been sequenced. The DNA sequence data along with the results of a detailed RNA analysis disclosed two unusual features of this green algal large subunit rRNA gene: (1) the presence of six group I introns (CeLSU.1-CeLSU.6) whose insertion positions have not been described previously, and (2) the presence of three short internal transcribed spacers that are post-transcriptionally excised to yield four rRNA species of 280, 52, 810 and 1720 nucleotides, positioned in this order (5' to 3') in the primary transcript. Together, these RNA species can assume a secondary structure that is almost identical to that proposed for the 23 S rRNA of Escherichia coli. All three internal transcribed spacers map to variable regions of primary sequence and/or potential secondary structure, whereas all six introns lie within

highly conserved regions. The first three introns are inserted within the sequence encoding the 810 nucleotide rRNA species and map within domain II of the large subunit rRNA structure; the remaining introns, found in the sequence encoding the 1720 nucleotide rRNA species, lie within either domain IV or V, as is the case for all other large subunit rDNA introns that have been documented to date. CeLSU.5 and CeLSU.6 each contain a long open reading frame (ORF) of more than 200 codons. While the CeLSU.6 ORF is not related to any known ORFs, the CeLSU.5 ORF belongs to a family of ORFs that have been identified in Podospora and Neurospora mitochondrial group I introns. The finding that a polymorphic marker showing unidirectional gene conversion during crosses between C. eugametos and Chlamydomonas moewusii is located within the CeLSU.5 ORF makes it likely that this intron is a mobile element and that its ORF encodes a site-specific endonuclease promoting the transfer of the intron DNA sequence.

77

Structural Similarities between psbA Genes from Red and Brown Algae

Winhauer, T.; Jager, S.; Valentin, K.; Zetsche, K.

Source: CURRENT GENETICS 20(1/2):177-180 (1991).

Descriptors: rhodophyta; phaeophyta; genes; plant proteins; photosystem ii; cloning; nucleotide sequences; plastids; chloroplast genetics; evolution; amino acid sequences; deletions

DNAL Call No.: QH426.C8

Abstract:

The single copy psbA genes from the multicellular red alga Antithamnion spec. and the brown alga Ectocarpus siliculosus have been cloned and sequenced and monocistronic transcripts have been detected. Both genes contain an insertion of 21 bp at the 3' end which was also found in cyanobacteria and which is absent in chloroplasts and the chlorophyll b-containing prochlorophyte Prochlorothrix hollandica. These findings are in agreement with the hypothesis of a polyphyletic origin of plastids. Plastids of red and brown algae appear to be closely related.

78

The Group IIB Intron from the Green Alga Scenedesmus obliquus Mitochondrion: Molecular Characterization of the In Vitro Splicing Products

Winkler, M. and Kuck, U.

Source: CURRENT GENETICS 20(6):495-502 (1991).

Descriptors: scenedesmus; mitochondrial dna; introns; alternative splicing; nucleotide sequences; restriction mapping; molecular conformation; ribosomal rna; genes; ribosomal dna

DNAL Call No.: QH426.C8

Abstract:

In the presence of high molar salt concentrations, the mitochondrial group IIB intron (rI1) from the green alga Scenedesmus obliquus is capable of splicing in vitro. After establishing the optimal conditions for RNA processing the in vitro splicing products were unequivocally identified in self-splicing experiments by Northern hybridization analysis employing 3'end-labelled RNAs or exon-and/or intron-specific probes. Finally, two trans-esterification products were identified by sequencing of the spliced RNA. From our data we conclude that the processing of group II introns from both algal and yeast mitochondria is preceded by identical consecutive trans-esterification steps. The predicted secondary and tertiary structure of intron rI1 of S. obliquus contains all the motifs necessary for optimal self-splicing and which are characteristic of other group IIB introns from different species.

79

Use of a Transposon with Luciferase as a Reporter to Identify Environmentally Responsive Genes in a

Cyanobacterium
Wolk, C.P.; Cai, Y.; Panoff, J.M.
Source: PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA 88(12):5355-5359 (1991).
Descriptors: anabaena; algae; cell differentiation; dna libraries; luciferase; mutants; nitrogen; nutrient deficiencies; temperature; transcription
DNAL Call No.: 500 N21P
Abstract:

Anabaena, a filamentous cyanobacterium, is of developmental interest because, when deprived of fixed nitrogen, it shows patterned differentiation of N2-fixing cells called heterocysts. To help elucidate its early responses to a decrease in nitrogen, we used a derivative of transposon Tn5 to generate transcriptional fusions of promoterless bacterial luciferase genes, luxAB, to the Anabaena genome. Genes that responded to removal of fixed nitrogen or to other environmental shifts by increased or decreased transcription were identified by monitoring the luminescence of colonies from transposon-generated libraries. The Tn5 derivative transposed in Anabaena at ca. 1-4 X 10-5 per cell and permitted high-resolution mapping of its position and orientation in the genome and facile cloning of contiguous genomic DNA.

80
Nostoc Commune UTEX 584 Gene Expressing Indole Phosphate Hydrolase Activity in Escherichia coli
Xie, W.Q.; Whitton, B.A.; Simon, J.W.; Jager, K.; Reed, D.; Potts, M.
Source: JOURNAL OF BACTERIOLOGY 171(2):708-713 (1989).
Descriptors: nostoc; genes; gene expression; hydrolases; indoles; enzyme activity; phospatases
DNAL Call No.: 448.3 J82
Abstract:

A gene encoding an enzyme capable of hydrolyzing indole phosphate was isolated from a recombinant gene library of Nostoc commune UTEX 584 DNA in lambda gt10. The gene (designated iph) is located on a 2.9-kilobase EcoRI restriction fragment and is present in a single copy in the genome of N. commune UTEX 584. The iph gene was expressed when the purified 2.9-kilobase DNA fragment, free of any vector sequences, was added to a cell-free coupled transcription-translation system. A polypeptide with an Mr of 74,000 was synthesized when the iph gene or different iph-vector DNA templates were expressed in vitro. When carried by different multicopy plasmids and phagemids (pMP005, pBH6, pB8) the cyanobacterial iph gene conferred an Iph+ phenotype upon various strains of Escherichia coli, including a phoA mutant. Hydrolysis of 5-bromo-4-chloro-3-indolyl phosphate was detected in recombinant E. coli strains grown in phosphate-rich medium, and the activity persisted in assay buffers that contained phosphate. In contrast, indole phosphate hydrolase activity only developed in cells of N. commune UTEX 584, when they were partially depleted of phosophorus, and the activity associated with these cells was suppressed partially by the addition of phosphate to assay buffers. Indole phosphate hydrolase activity was detected in periplasmic extracts from E. coli (Iph+) transformants.

81
Repetitive Sequence-Mediated Rearrangements in Chlorella ellipsoidea Chloroplast DNA: Completion of Nucleotide Sequence of the Large Inverted Repeat
Yamada, T.
Source: CURRENT GENETICS 19(2):139-147 (1991).
Descriptors: chlorella ellipsoidea; repetitive dna; chloroplasts; nucleotide sequences; restriction mapping; amino acid sequences; genes; ribosomal rna; ribosomal dna; gene mapping; transfer rna
DNAL Call No.: QH426.C8

Abstract:

A 3454 base pair (bp) sequence of the large inverted repeat (IR) of chloroplast DNA (cpDNA) from the unicellular green alga Chlorella ellipsoidea has been determined. The sequence includes: (1) the boundaries between the IR and the large single copy (LSC) and the small single copy (SSC) regions, (2) the gene for psbA and (3) an approximately 1.0 kbp region between psbA and the rRNA genes which contains a variety of short dispersed repeats. The total size of the Chlorella IR was determined to be 15 243 bp. The junction between the IR and the small single copy region is located close to the putative promoter of the rRNA operon (906 bp upstream of the -35 sequence on each IR). The junction between the IR and the large single copy region is also just upstream of the putative psbA promoter, 218 bp upstream from the ATG initation codon. A few sets of unique sequences were found repeatedly around both junctions. Some of the sequences flanking the IR-LSC junction suggest a unidirectional and serial expansion of the IR within the genome. The psbA gene is located close to the LSC-side junction and codes for a protein of 352 amino acid residues. A highly conserved C-terminal Gly is absent. Unlike the psbA of Chlamydomonas species, which contains 2-4 large introns, the gene of Chlorella has no introns. The overall gene organization of the Chlorella IR is very different from that of higher plants, but a similar gene cluster of rrn-psbA is also found in the IR of Chlamydomonas species and in a single copy region of some chlorophyll a/c-containing algae, indicating a common evolutionary lineage of these cpDNAs. The origin and evolution of the IR structure are discussed in the light of these observations.

82

Sequences of trnR-ACG and petD that Contain a tRNA-like Element within the Chloroplast Genome of Chlamydomonas reinhardtii

Yu, W. and Spreitzer, R.J.

Source: NUCLEIC ACIDS RESEARCH 19(4):957 (1991).

Descriptors: chlamydomonas reinhardtii; chloroplast genetics; transfer rna; nucleotide sequences

DNAL Call No.: QD341.A2N8

Products and Product Development

83

Glycerol

Agarwal, G.P.

Source: ADVANCES IN BIOCHEMICAL ENGINEERING/BIOTECHNOLOGY (41):95-128 (1990).

Descriptors: saccharomyces cerevisiae; yeasts; endomycetales; algae; glycerol; biosynthesis; biotechnology; literature review

DNAL Call No.: TP248.3.A38

Abstract:

Glycerol is traditionally produced as a by-product of soap and fatty acids industries. The demand for glycerol has always exceeded the supply from these industries so the excess demand has been met by chemical synthesis from propylene for the last several decades. Though glycerol production has a long history (dating back to World War I) of being produced via a biochemical route, yet it is not sufficiently developed to compete with the chemical route. In the present review a case has been made to produce glycerol via any of the several known biochemical routes: a) Sulfite-Alkali-Steered Yeast Process b)

Bacterial Process c) Osmotolerant Yeast Process d) Algal Cultivation Process. The possible reasons for these processes not being able to compete with chemical processes are critically reviewed. The literature on downstream processing of any of the biochemical processes is quite limited and more investigations are required into this aspect to make these processes viable. The biosynthesis mechanism of glycerol production in the organisms is summarized and the need to look into some of the fundamental aspects of glycerol synthesis in an osmotolerant yeast emphasized. The comparison between the various processes is made wherever possible.

84

Enriching Marine Macroalgae with Eicosatetraenoic Arachidonic and Eicosapentaenoic Acids by Chilling

Al-Hasan, R.H.; Hantash, F.M. Radawan, S.S.

Source: APPLIED MICROBIOLOGY AND BIOTECHNOLOGY 35(4):530-535 (1991).

Descriptors: algae; methods; lipids; fatty acids; biotechnology industry

DNAL Call No.: QR1.E9

Abstract:

Twelve macroalgae belonging to the Chlorophyta, Phaeophyta and Rhodophyta were collected from the Arabian Gulf. Field samples and samples that were first incubated at 5.degree. C and 24.degree. C in the light for 1 week were analysed for lipids and fatty acids. The lipid contents varied according to the macroalga and, within the Chlorophyta and Phaeophyta, some representatives accumulated more lipids at 5.degree. C and others at 24.degree. C. All samples of algae had similar lipid composition with only quantitative differences. The temperature did not have a common effect on the lipid composition of representative algae, although changes in the relative concentration of specific classes were recorded. The Phaeophyta and Rhodophyta were as a rule richer than the Chlorophyta in eicosatetraenoic (20:4) and eicosapentaenoic (20:5) but poorer in linolenic (18:3) acids. In most of the algae, incubation at 5.degree. C was associated with lowering the proportion of palmitic acid (16:0) in the total lipids, and, but only in the Phaeophyta and Rhodophyta, increasing the concentration of 20:4 and 20:5. These polyunsaturated fatty acids occurred in high levels in monogalactosyldiacylglycerols (MGDG) and digalactosyldiacylglycerols (DGDG) of the Phaeophyta and Rhodophyta but not the Chlorophyta, the MGDG and DGDG of which were rich in 18:3 and hexadecatrienoic acid (16:3).

85

Biosynthesis of 130-Kilodalton Mosquito Larvicide in the Cyanobacterium Agmenellum-quadruplicatum PR-6

Angsuthanasombat, C. and Panyim, S.

Source: APPLIED AND ENVIRONMENTAL MICROBIOLOGY

Descriptors: bacillus thuringiensis; genes; genetic transformation; cyanobacteria; ovicides and larvicides; biological control; diptera

DNAL Call No.: 448.3 AP5

Abstract:

The 130 kilodalton mosquito larvidical gene, cloned from Bacillus thuringiensis var. israelensis, was introduced into the cyanobacterium Agmenellum quadruplicatum PR-6 by plasmid transformation. Transformed cells synthesized 130-kilodalton delta-endotoxin protein and showed mosquito larvicidal activity. Results demonstrate a potential use of a cyanobacterium for biological control of mosquitoes.

86

Anaerobic Digestion of Seaweed for Biogas a Kinetic Evaluation

Anjaneyulu, K.; Tarwadi, S.J.; Mehta, D.J.
Source: JOURNAL OF CHEMICAL TECHNOLOGY AND BIOTECHNOLOGY 45(1):5-14 (1989).
Descriptors: sargassum-tenerrimum; fermentation; biotechnology industry
DNAL Call No.: QR53.J685
Abstract:

A kinetic study of biogas production in batch digesters by anaerobic digestion of seaweed, Sargassum tenerrimum, with a mixed bacterial culture consisting of methanogenic bacteria and an algin-degrading bacterial strain was carried out at different concentrations of dry total solids. Specific rate constants of biogas production during the lag, exponential and monomolecular (stationary) phases of bacterial growth were determined. About half the total volume of biogas was generated during the exponential phase irrespective of the concentration of seaweed in the digesters. The specific rates of substrate destruction and biogas generation in the stationary phase decreased with increasing substrate concentration. The yield of biogas per gram dry total solids of seaweed was about the same at all concentrations, but with a marked decline at 12% (w/v) total solids. The maximum destruction of volatile solids effected was about 63% over a period of 72 days.

87
Agar and Agarose Biotechnological Applications
Armisen, R.
Source: HYDROBIOLOGIA 221(0):157-166 (1991).
Descriptors: agar; agarose; bacteriological agar; chromatography; electrophoresis
DNAL Call No.: 410 H992

88
Biotechnology for Rural Nutrition an Economic Evaluation of Algal Protein Supplements in South India
Babu, S.C. and Rajasejaren, B.
Source: FOOD POLICY 16(5):405-414 (1991).
Descriptors: algae; human; developing countries; food policy; costs
DNAL Call No.: HD9000.1.F66
Abstract:

This article evaluates the introduction of algal biotechnology as a nutrition intervention in rural south India in terms of its benefits, costs and acceptability to the tastes and income of rural households. Data were collected from a whole-village study, and developments in the theory of the economics of tastes are used to analyze the socioeconomic acceptance of algal supplements among the study households. The results indicate that the algal food supplement is highly cost effective in providing adequate protein and other micronutrients. However, the absolute deviation between actual and optimal intake of algal food increased with higher income class. Several policy implications for food and nutrition interventions in developing countries are derived.

89
A Marketing Approach to Agar
Becker, K.J. and Rotmann, K.W.G.
Source: JOURNAL OF APPLIED PHYCOLOGY 2(2):105-110 (1990).
Descriptors: consumer awareness; thickener; laxative; packaging; biotechnology; far east; japan; germany; islamic countries
DNAL Call No.: QK564.J68
Abstract:

Agar has, with the exception of certain retail markets in the Far East, specifically Japan,

traditionally been sold to the industrial user. Small quantities are consumed in Islamic countries during Ramadan and in the Germanic countries as a food thickener and a laxative. However, outside of Japan, no significant marketing effort has ever been undertaken with a view to increase the demand for agar by consumers. A marketing plan is suggested to change this situation. All possible uses for agar by the consumer have been identified and studied. The special features of the product, together with certain packaging, are highlighted. Potential markets for these features are identified. Strategies for the development of these markets have been developed. The overall plan is now in a state of final review and just prior to implementation. The product launch should generate a significant consumer awareness which will translate into demand, thereby increasing the market for agar in various forms, formulations and packagings.

90

The Biotechnology of Cultivating Dunaliella for Production of Beta-Carotene Rich Algae

Ben-Amotz, A.; Shaish, A.; Avron, M.

Source: BIORESOURCE TECHNOLOGY 38(2/3):233-235 (1991).

Descriptors: dunaliella; algae culture; beta-carotene; biosynthesis; light intensity; growth inhibitors; isomerization; phytoene; food biotechnology; mass cultivation; salt tolerance; glycerol

DNAL Call No.: TD930.A32

Abstract:

Dunaliella accumulates massive amounts of .beta.-carotene when cultivated under high light intensity and growth-limiting conditions. The pathway for biosynthesis of .beta.-carotene was elucidated by analysis of the effect of selected inhibitors. The presence of the inhibitors elicited the accumulation of the following intermediates: .beta.-zeacarotene, lycopene, .zeta.-carotene, phytofluene, phytoene and a few unidentified long-chain isoprenoids. Each of the accumulated intermediates was composed of about equal amounts of two stereoisomers, as is the case for .beta.-carotene in the untreated algae. It is deduced, therefore, that the isomerization reaction occurs early in the pathway of .beta.-carotene biosynthesis, at or before phytoene. The unique carotenogenesis properties of Dunaliella led to the development of a new biotechnological process for mass-cultivation of the alga. Commercial production facilities for .beta.-carotene rich Dunaliella exist today in Israel, USA, Australia, Spain and China. Recent developments, which indicate that the stereoisomeric mixture of .beta.-carotene present in Dunaliella is preferentially absorbed in animal tissues, coupled with new evidence for the efficacy of .beta.-carotene in reducing the incidence of cancer, have opened new vistas of potential markets for the high .beta.-carotene algae.

91

Microbial Production of Hydrocarbons

Birch, L.D. and Bachofen, R.

Source: BIOTECHNOLOGY: A COMPREHENSIVE TREATISE: BIOTECHNOLOGY, VOL.6B. SPECIAL MICROBIAL PROCESSES. Rehm, H.J., editor. VCH Publishers, Inc., New York, 1989, pp.71-100.

Descriptors: review; algae; cyanobacteria; bacteria; trichomonads; hydrogenases; nitrogenases; formate; hydrogenylase; biotechnology

DNAL Call No.: QR53.B52

92

Elucidation and Optimization of the Medium Constituents Controlling Antibiotic Production by the Cyanobacterium Nostoc-muscorum

Bloor, S. and England, R.R.

Source: ENZYME AND MICROBIAL TECHNOLOGY 13(1):76-81 (1991).

Descriptors: algae; nitrate; iron; antibiotics; biotechnology industry; pharmaceutical industry
DNAL Call No.: TP248.E5E565
Abstract:

A study has been made to determine which nutrient factors control antibiotic production by the cyanobacterium, Nostoc muscorum. A two-phase approach was employed using a factorial method to explore the response surface and a steepest ascent method to climb the response surface to the region of the optimum. It was found that nitrate and iron were the factors significantly affecting antibiotic production; 26.4 mM nitrate and 6.mu.M iron were the optimal concentrations for maximizing antibiotic production by N. muscorum.

93
Development of Western Biotechnology's Algal Beta-carotene Plant
Borowitzka, L.J.
Source: BIORESOURCE TECHNOLOGY 38(2/3):251-252 (1991).
Descriptors: algae culture; dunaliella; industrial methods; beta-carotene; food colorants
DNAL Call No.: TD930.A32
Abstract:

In the past ten years, laboratory studies and open pond experiments at Hutt Lagoon, in Western Australia, have developed a commercial process, for extracting the food colouring, .beta.-carotene, from algal cultures. The hypersaline microscopic alga, Dunaliella salina, is grown in 50 ha of open ponds, harvested, and the .beta.-carotene concentrated and packaged as 2% and 20% suspensions in vegetable oil.

94
Algal Biotechnology Products and Processes Matching Science and Economics
Borowitzka, M.A.
Source: JOURNAL OF PHYCOLOGY (3 suppl.):10 (1991).
Descriptors: abstract; carotenoids; beta carotene; phycocyanin; culture systems; harvesting processing
DNAL Call No.: QK564.J6

95
Hydrogen Production by Eukaryotic Algae
Brand, J.J.; Wright, J.N.; Lien, S.
Source: BIOTECHNOLOGY AND BIOENGINEERING 33(11):1482-1488 (1989).
Descriptors: fermentation; biotechnology industry
DNAL Call No.: 381 J8224

96
Environmental Control of Lipid and Biomass Production in Two Diatom Species
Chelf, P.
Source: J APPL PHYCOL 2(2):121-130 (1990).
Descriptors: chaetoceros-muelleri-var-subsalsum; navicula-saprophila; nitrogen biotechnology
DNAL Call No.: QK564.J68
Abstract:

Biomass and neutral lipid accumulation were examined in Chaetoceros muelleri var. subsalsum and Navicula saprophila using a multivariate, fractional factorial design. Variables included were conductivity, temperature, nitrogen concentration, silicon concentration, time (culture age), and alkalinity. Measured characteristics included nile red fluorescence (as a measure of neutral lipid content) and ash-free dry weight (AFDW). Nitrogen concentration was the variable with the greatest effect on neutral lipid and ash-

free dry weight accumulation over the ranges tested. Increasing conductivity in the range examined had a significant, negative impact on neutral lipid accumulation in both of these strains, while increasing alkalinity had a positive effect on lipid and ash-free dry weight in both strains. Experimental designs such as those described here have great potential utility in biological systems with complex interactions.

97

Biosynthesis of High Concentrations of an Exopolysaccharide during the Cultivation of the Microalga Botryococcus-braunii

Fernandes, H.L.; Tome, M.M.; Lupe, F.M.; Fialho, A.M.; Sa-Correia, I.; Novais, J.M.

Source: BIOTECHNOLOGY LETTERS 11(6):433-436 (1989).

Descriptors: fermentation; biotechnology industry

DNAL Call No.: QR53.B56

Abstract:

A non-axenic strain of the microalga Botryococcus braunii Kutzing, isolated from a small lake in Portugal, when cultured at 25.degree.C in mineral medium and under continuous illumination, showed a poor production of hydrocarbons (5% of the dry biomass) but excreted remarkably high quantities of an exopolysaccharide (4-4.5 g/l) into the medium. The production of soluble polysaccharide with galactose, fucose and uronic acid residues, follows growth. The role of the mucoid contaminating bacteria in polysaccharide production in the mixed culture was unproven.

98

Analysis of the Biomass Quality and Photosynthetic Efficiency of a Nitrogen-Fixing Cyanobacterium Grown Outdoors with Two Agitation Systems

Fontes, A.G.; Moreno, J.; Vargas, M.A.

Source: BIOTECHNOLOGY AND BIOENGINEERING 34(6):819-824 (1989).

Descriptors: biotechnology industry; airlift; paddlewheel; productivity; protein; energy conversion efficiency; nitrogen fixation

DNAL Call No.: 381 J8224

Abstract:

The efficiency of two different agitation systems (airlift and paddlewheel) in the biomass photoproduction of a nitrogen-fixing filamentous blue-green alga was evaluated outdoors, and the elemental and molecular composition of the cells grown with each system was analyzed. With the paddlewheel system, the productivity values achieved were over 30% higher than with the airlift system, both in summer and winter. In this last season, a conversion efficiency of total solar energy into stored biomass energy of 3.3% was estimated for the paddlewheel system. Moreover, the algal cells grown with this system exhibited a higher net protein (58.9% of dry weight) and nitrogen (11.3%) content than those grown with the airlift device, with an estimated nitrogen fixation rate of more than 2 g N m-2 day-1. These advantages of the paddlewheel system make this procedure more appropriate for the large-scale photoproduction of nitrogen-fixing blue-green algae outdoors.

99

Effect of Low-dose Ultrasonic Treatment on Physiological Variables in Anabaena Flos-aquae and Selenastrum-capricornutum

Francko, D.A.; Taylor, S.R.; Thomas, B.J.; McIntosh, D.

Source: BIOTECHNOLOGY LETTERS 12(3):219-224 (1990).

Descriptors: anabaena flos-aquae; algae; ultrasonics; biomass accumulation; chlorophyll; alkaline phosphatase; growth rate; biotechnology

DNAL Call No.: QR53.B56

Abstract:

Cell protein content in two species of cultured algae Anabaena flos-aguae and Selenastrum capricornutum, was markedly enhanced by low-dose, short-duration ultrasonic treatment. Chlorophyll a levels and 14C-bicarbonate uptake rates were not affected by ultrasonic treatment in either species. Ultrasonically-activated Anabaena cultures placed in media deficient in nitrogen and phosphorus produced more biomass per unit time, exhibited less cell-surface alkaline phosphatase activity per cell, and had a higher heterocyst frequency than non-sonicated, nutrient-deficient cultures. In contrast, sonicated, nutrient-deficient Selenastrum cultures grew more slowly and had higher alkaline phosphatase activity than non-sonicated variants. Collectively, the data support that key metabolic variables may be altered by ultrasonic treatment in algal cultures and that the magnitude and direction of change may be species specific.

99

Hydrocarbon Recovery and Biocompatibility of Solvents for Extraction from Cultures of Botryococcus-braunii

Frenz, J.; Largeau, C.; Casadevall, E.; Kollerup, F.; Daugulis, A.J.

Source: BIOTECHNOLOGY AND BIOENGINEERING 34(6):754-762 (1989).

Descriptors: fermentation; biotechnology; yield; cell viability; cell wall; chromatography

DNAL Call No.: 381 J8224

Abstract:

Various water-immiscible solvents were tested for biocompatibility and hydrocarbon recovery under different contact conditions with the hydrocarbon-rich microalga Botryococcus braunii. Eighteen solvents were first selected from a data base of 1500 compounds (compiled for solvent selection for ethanol recovery from Saccharomyces cerevisiae fermentation). Nine of these candidate solvents were shown to be biocompatible with B. braunii following short contact times. This biocompatibility tends to be associated with high molecular weights and high boiling points but strongly depends on solvent chemical structure. A low polarity is essential to biocompatibility and calculated octanol-water partition coefficients, or capacity factors determined by reversed-phase high-performance liquid chromatography (HPLC), are suitable predictors of biocompatibility with B. braunii. High recoveries of hydrocarbons directly from the algal culture require relatively polar solvents and are, therefore, inimical with maintenancee of cell viability. The inaccessibility of weakly polar solvents to the cell surface appears to protect the algae but also prevents substantial recovery of the hydrocarbons stored in B. braunii outer walls. In order to achieve a high recovery, contact with the solvent must be carried out on algae concentrated by filtration. Then, a large fraction of B. braunii hydrocarbons can be recovered, after a short contact time, without impairing cell viability. Under these conditions, the pertinent solvent property is affinity for the nonpolar hydrocarbons, and the highest recovery yield, .apprx. 70% after contact for 30 min, is achieved with hexane.

100

Hydrocarbon Recovery by Extraction with a Biocompatible Solvent Form Free and Immobilized Cultures of Botryococcus-braunii

Frenz, J.; Largeau, C.; Casadevall, E.

Source: ENZYME AND MICROBIAL TECHNOLOGY 11(11):717-724 (1989).

Descriptors: algae; biotechnology industry; growth; bioreactor; colony size; alginate beads; yield

DNAL Call No.: TP248.E5E565

Abstract:

Recovery of a substantial fraction of B. braunii hydrocarbons was achieved via short

contact with hexane of algae concentrated by filtration. Growth and hydrocarbon production during subsequent cultures were not impaired, even after repeated extractions. In fact, the hydrocarbon content of the cultures derived from treated algae tends to be higher than in controls. Recovery yields can be influenced by the physiological stage of the extracted culture. In addition, algae corresponding to the early exponential stage afford higher recoveries when grown under air-lift conditions relative to standard conditions; this likely originates from the smaller average size of colonies in the former cultures. The scale-up of extraction indicates that the recovery yield falls off when relatively large amounts of algae are contacted with hexane (large clump formation due to sharp polarity contrast between wet cells and the nonpolar solvent). Immobilization, via entrapment in alginate beads and adsorption on polyurethane foams, was used to overcome this problem. Contact with hexane does not affect subsequent growth and hydrocarbon production of immobilized cultures. Recovery yields are markedly increased, relative to free cells, especially in the case of polyurethane foams.

101
Production of Bioflavor by Regeneration from Protoplasts of Ulva-pertusa ulvales Chlorophyta
Fujimura, T. and Kajiwara, T.
Source: HYDROBIOLOGIA 204-205(0):143-150 (1990).
Descriptors: cappa carrageenan; agarose; agar; polymers; long chain aldehydes; marine biotechnology
DNAL Call No.: 410 H992

102
Actual Potential and Speculative Applications of Seaweed Cellular Biotechnology some Specific Comments of Gelidium
Garcia-Reina, G.; Gomez-Pinchetti, J.L.; Robledo, D.R.; Sosa, P.
Source: HYDROBIOLOGIA 221(0):181-194 (1991).
Descriptors: callus; cell culture; domestication; protoplast; tissue culture; organogenesis; method; application
DNAL Call No.: 410 H992

103
Growth and Nitrogen Fixation by Immobilized Cyanobacteria
Gendel, S.M. and Nohr, R.S.
Source: APPLIED MICROBIOLOGY AND BIOTECHNOLOGY 31(2):138-145 (1989).
Descriptors: nostoc muscorum; nitrogen fixation; photosynthesis; growth rate; immobilization
DNAL Call No.: QR1.E9
Abstract:

The nitrogen-fixing photosynthetic cyanobacteria have significant potential for utilization as a biological system for the production of reduced nitrogen compounds, either by industrial fermentation or in the environment as soil inocula. In either system, the ability to immobilize cyanobacteria on the external surface of fibrous substrata would significantly improve the ease of manipulation of the cells, control of growth, and product recovery without the complications inherent in immobilization by entrapment. We have shown that the filamentous heterocystous species Nostoc muscorum is naturally able to attach to a variety of different fibres, both natural and artificial. Attached cells are able to grow and fix nitrogen in both liquid and plate culture. Nitrogen-fixing cells attach to the fibres much more readily than do non-fixing cells, suggesting that the physiological and morphological changes accompanying heterocyst differentiation result in the production of specific attachment sites. Scanning electron microscopy of attached cells shows that heterocysts act as attachment sites and that the external cell wall material specifically

synthesized around the heterocysts may be acting as the biological "glue" for this attachment.

104
Biotechnological Principles for Hydrogen Production by Phototrophic Microorganisms
Gogotov, I.N.
Source: SOVIET BIOTECHNOLOGY (1):11-16 (1989).
Descriptors: hydrogen; production; phototropism; microorganisms; anaerobiosis; aerobiosis; cyanobacteria
DNAL Call No.: TP248.13.S68

105
Microbial Formation of Manganese Oxides
Greene, A.C. and Madgwick, J.C.
Source: APPLIED AND ENVIRONMENTAL MICROBIOLOGY 57(4):1114-1120 (1991).
Descriptors: chlamydomonas; pseudomonas; manganese oxides; manganese dioxide; manganese; oxidation; chemical precipitation; industrial applications; industrial microbiology; bioreactors; biotechnology
DNAL Call No.: 448.3 AP5
Abstract:

Microbial manganese oxidation was demonstrated at high Mn2+ concentrations (5 g/liter) in bacterial cultures in the presence of microalga. The structure of the oxide produced varied depending on the bacterial strain and mode of culture. A nonaxenic, acid-tolerant microalga, a Chlamydomonas sp., was found to mediate formation of manganite (.gamma.-MnOOH). Bacteria isolated from associations with crude cultures of this alga grown in aerated bioreactors formed disordered .gamma.-MnO2 from Mn2+ at concentrations of 5 g/liter over 1 month, yielding 3.3 g of a semipure oxide per liter. All algal-bacterial cultures removed Mn2+ from solution, but only those with the highest removal rates formed an insoluble oxide. While the alga was an essential component of the reaction, a Pseudomonas sp. was found to be primarily responsible for the formation of a manganese precipitate. Medium components.sbd.algal biomass and urea.sbd.showed optima at 5.7 and 10 g/liters, respectively. The scaled-up culture (50 times) gave a yield of 22.3 g (53 mg/liter/day from a 15-liter culture) of semipure disordered .gamma.-MnO2, identified by X-ray diffraction and Fourier transform infrared (FTIR) spectroscopy, and had a manganese oxide O/Mn ratio of 1.92. The Mn(IV) content in the oxide was low (30.5%) compared with that of mined or chemically formed .gamma.-MnO2 (ca. 50%). The shortfall in the bacterial oxide manganese content was due to biological and inorganic contaminants. FTIR spectroscopy, transmission electron microscopy, and electron diffraction studies have identified manganite as a likely intermediate product in the formation of disordered .gamma.-MnO2.

106
The Influence of Light-Dark Cycles in Mixed Algal Cultures on their Productivity
Grobbelaar, J.U.
Source: BIORESOURCE TECHNOLOGY 38(2-3):189-194 (1991).
Descriptors: algae; aquaculture; energy; yield; environment; cycles; oxygen liberation; biotechnology industry
DNAL Call No.: TD930.A32
Abstract:

In mass algal cultures, some form of agitation is usually provided, which amongst others, moves the organisms through an optically dense profile. During this transport, fluctuations in the light energy supply are perceived by the algae, which are of the order

of 1 Hz and less. Additional to these variations the cultures are subject to diurnal, seasonal and climatic light variations. It has been suggested that turbulence with the resultant light/dark cycles enhances their productivity. However, turbulence has two major influences on an organism, i.e. it facilitates flucutating light regimes and decreases the boundary layer which results in an increased exchange rate between the organism and its environment. With the aid of oxygen liberation measurements, the influence of fluctuating light regimes on productivity was measured. No simple relation existed, but no enhancement of productivity could be shown at cycles of 1-0.0038 Hz. Short term physiological changes were found to influence productivity severely.

107

Preparation of Protoplasts from the Carrageenophyte Gigartina corymbifera (Kutz.) J. Ag. (Rhodophyta)

Gross, W.

Source: JOURNAL OF MICROBIOLOGICAL METHODS 12(3/4):217-233 (1990).

Descriptors: rhodophyta; protoplasts; viability; isolation techniques; carrageenan; enzymes; mixtures; pseudomonas; cell walls; genetic engineering; genetic improvement

DNAL Call No.: QR65.J68

Abstract:

Protoplasts were isolated with high yield from the carrageenophyte Gigartina corymbifera (Kutz.) J. Ag. by using the enzyme carrageenase in combination with cell wall-digesting enzymes. The enzyme mixture consisted of 5 U carrageenase.ml-1, 2% cellulase, 2% Macerozyme R-10, and 0.2% Pectolyase Y-23 dissolved in 60% seawater containing 0.7 M mannitol, 5mM CaCl2, and 40 mM Tris-HCL, pH 7. Carrageenase was prepared from cultures of the marine bacterium Pseudomonas carrageenovora. Protoplasts from G. corymbifera were spherical, dark red-colored and very uniform is size (approximately 17 micrometer); they originated solely from the epidermal tissue and > 95% of the protoplasts were viable. Freshly prepared protoplasts lack cell walls and polysaccharide sheaths, as demonstrated by electron microscopy and specific staining methods. Spontaneous cell fusion was observed on several occasions. These protoplasts could serve as a useful tool in crop improvement of this important carrageenan-producing alga.

108

Chemical and Physical Properties of Algal Polysaccharides used for Cell Immobilization

Guiseley, K.B.

Source: ENZYME AND MICROBIAL TECHNOLOGY 11(11):706-716 (1989).

Descriptors: review; microorganisms; enzymes; biotechnology industry; crosslinking; algin; agar; agarose; carrageenan

DNAL Call No.: TP248.E5E565

109

Automatic On-line Growth Estimation Method for Outdoor Algal Biomass Production

Guterman, H.; Ben-Yaakov, S.; Vonshak, A.

Source: BIOTECHNOLOGY AND BIOENGINEERING 34(2):143-152 (1989).

Descriptors: oxygen; production rate; photosynthetic respiration; industrial application; wastewater treatment; automation; biotechnology industry

DNAL Call No.: 381 J8224

Abstract:

An on-line measuring procedure for estimating productivity in outdoor algal cultures was developed and tested experimentally. The procedure is based on a previously described method for on-line measuring net O2 production rate (OPR). The data obtained by this method was found to correlate well with the conventional procedures for estimation

productivity by measuring the changes in biomass concentration in the culture. The new procedure seems to be superior to the latter since it can be carried out in an almost continuous way and can give immediate indication on the productivity. OPR could be used to monitor on-line the photosynthetic and/or respiration activity in small research fermentors or in large-scale open systems outdoors.

110
Gamma Linolenic Acid Production by Microalgae
Hirano, M.; Mori, H.; Miura, Y.; Matsunaga, N.; Nakamura, N.; Matsunaga, T.
Source: APPLIED BIOCHEMISTRY AND BIOTECHNOLOGY 24-25(spring-summer):183-192 (1990).
Descriptors: spirulina-platensis; fermentation biotechnology
DNAL Call No.: QD415.A1J62

111
Effect of Light and Carbon Dioxide on Biopolymer Production by the Unicellular Red Alga Porphyridium-Cruentum
Iqbal, M.; Stepan-Sarkissian, G.; Grey, D.; Fowler, M.W.
Source: FOOD BIOTECHNOLOGY 4(1):104 (1990).
Descriptors: abstract; algae; food industry use; biotechnology
DNAL Call No.: TP248.65.F66F66

112
Screening Test for Deodorizing Substance from Marine Algae and Identification of Phlorotannins as the Effective Ingredients in Eisenia-bicyclis
Kita, N.; Fujimoto, K.; Nakajima, I.; Hayashi, R.; Shibuya, K.
Source: JOURNAL OF APPLIED PHYCOLOGY 2(2):155-162 (1990).
Descriptors: eisenia-bicyclis; ecklonia-cava; ecklonia-kurome; mercaptan; trapping effect; chlorophyll; sodium; copper; chlorophyllin; biotechnology
DNAL Call No.: QK564.J68
Abstract:

Aqueous extracts from 33 species of marine algae were assessed for their methyl mercaptan-trapping activity by gas chromatography to search for novel natural oral deodorants. Brown algae belonging to the Laminariales such as Eisenia bicyclis, Ecklonia cava and Ecklonia kurome were found to show remarkable deodorizing action against methyl mercaptan. The effective components in Eisenia bicyclis were identified as a phlorotannin, a group of molecules which are characteristic components of Laminariales. In addition phlorotannins extracted from E. bicyclis were more effective at reducing methyl mercaptan than conventional natural deodorants such as chlorophyll and sodium copper chlorophyllin.

113
Microbial Production of Hydrogen
Kosaric, N. and Lyng, R.P.
Source: BIOTECHNOLOGY: A COMPREHENSIVE TREATISE: BIOTECHNOLOGY, VOL. 6B. SPECIAL MICROBIAL PROCESSES. Rehm, H.J., editor. VCH Publishers, Inc., New York, 1989, pp.102-134.
Descriptors: review; algae; cyanobacteria; bacteria; trichomonads; hydrogenases; nitrogenases; formate; hydrogenylase; biotechnology
DNAL Call No.: QR53.B52

114

Macrokinetics and Mathematical Modelling of Quinone Reduction by Cyanobacteria

Kreysa, G. and Kraemer, P.

Source: JOURNAL OF CHEMICAL TECHNOLOGY AND BIOTECHNOLOGY 44(3):205-218 (1989).

Descriptors: anabaena; anacystis-nidulans; bioelectrochemical fuel

DNAL Call No.: QR53.J685

Abstract:

The kinetics of the reduction of externally added 2-hydroxy-1,4-naphthoquinone by blue-green algae of the strains Anabaena PCC 7120 and Anacystis nidulans PCC 6301 were studied in aqueous cell suspensions by electrochemical monitoring of the concentration of the formed hydroquinone. This reaction is of potential interest for bioelectrochemical fuel cells. The experimental curves obtained could be interpreted by a model that takes into account that both substrate and product have to be transported through the microbial cell walls and that the conversion reaction takes place with first-order kinetics within the microbial cells. No clear evidence was found for the involvement of photosynthesis. It is suggested that the reduction of the quinone probably occurs via the enzyme catalyzed oxidation of endogenous storage product(s), presumably glycogen.

115

Market Applications for Microalgae

Kyle, D.

Source: JOURNAL OF THE AMERICAN OIL CHEMISTS' SOCIETY 66(5):648-651

Descriptors: algae; microorganisms; industrial applications; oils and fats industry; food biotechnology; algae culture

DNAL Call No.: 307.8 J82

116

A Study of the Energetics and Economics of Microalgal Mass Culture with the Marine Chlorophyte Tetraselmis-suecica Implications for Use of Power Plant Stack Gases

Laws, E.A. and Berning, J.L.

Source: BIOTECHNOLOGY AND BIOENGINEERING 37(10):936-947 (1991).

Descriptors: algae; biotechnology industry; air pollution control; carbon dioxide; natural energy laboratory Hawaii USA

DNAL Call No.: 381 J8224

Abstract:

The marine phytoplankter Tetraselmis suecica was grown in shallow outdoor flumes for a period of approximately 6 months at the Natural Energy Laboratory of Hawaii. In full sunlight, gross production rates were 15-20 g C m-2 d-1. The corresponding photosynthetic efficiencies (PE's) were 9-10%. Respiration losses removed about half the gross production. The CO2 utilization efficiencies of 96 .+-. 11% were achieved by bubbling CO2 into the culture with the use of a counterflow sump system. Adding the CO2 in the form of carbonated water resulted in utilization efficiencies of 81 .+-. 11%. Archimedes screws proved superior to both paddle wheels and propellers as a means of circulating the water in the flumes. Insertion of foil arrays into the flumes to effect systematic mixing of the culture significantly enhanced production. The enhancement was greater when the foils were oriented at a small angle relative to the horizontal than when they were oriented at the same angle relative to the vertical. Light modulation effect are implicated as the probable cause of most of the enhancement. Substitution of electric power plant stack gases for pure CO2 resulted in no significant change in the production of T. suecica grown in chemostat culture.

117
Protoplast Production in Chondrus-crispus Gametophytes Gigartinales Rhodophyta
Le Gall, Y.; Braud, J.P.; Kloareg, B.
Source: PLANT CELL REPORTS 8(10):582-585 (1990).
Descriptors: marine bacteria; cellulase; carrageenase; algae regeneration; cell culture; yield; crop industry; biotechnology industry
DNAL Call No.: QK725.P54
Abstract:

Protoplasts were isolated from female gametophytes of Chondrus crispus (Stackh.) using commercial cellulase and various carrageenases prepared from marine bacteria. Depending on the nature of the donor tissue (apices or whole thallus, wild or cultivated strains), yields ranged from 1.0-8.5 .times. 108 protoplasts per gram of fresh tissue. Preincubating the tissue with a potassium chelator, Kryptofix 222, enhanced protoplast yields by 30-50%. based on staining with fluorescein diacetate most protoplasts were viable. A few protoplasts regenerated a cell wall and divided.

118
Glutamate Production from Carbon Dioxide by Marine Cyanobacterium Synechococcus-sp Using a Novel Biosolar Reactor Employing Light-Diffusing Optical Fibers
Matsunaga, T.; Takeyama, H.; Sudo, H.; Oyama, N.; Ariura, S.; Takano, H.; Hirano, M.; Burgess, J.G.; Sode, K.; Nakamura, N.
Source: APPLIED BIOCHEMISTRY AND BIOTECHNOLOGY 28-29 (0):157-168 (1991).
Descriptors: biotechnology
DNAL Call No.: QD415.A1J62

119
Opportunities and Applications of Biotechnology in the Food Industry
Meek, S.D.
Source: FOOD AUSTRALIA-OFFICIAL JOURNAL OF CAFTA AND AIFST 42(1):12-13 (1990).
Descriptors: food industry; biotechnology; food quality; ingredients; food processing; cheeses; algal cultures; patents; secondary metabolites; food wastes
DNAL Call No.: TP368.F662

120
Lipids and Macromolecular Lipids of the Hydrocarbon-Rich Microalga Botryococcus braunii. Chemical Structure and Biosynthesis. Geochemical and Biotechnological Importance.
Metzger, P.; Largeau, C.; Casadevall, E.
Source: FORTSCHRITTE DER CHEMIE ORGANISCHER NATURSTOFFE=PROGRESS IN THE CHEMISTRY OF ORGANIC NATURAL PRODUCTS 57:1-70 (1991).
Descriptors: algae; lipogenesis; hydrocarbons; fatty acids; triacylglycerols; sterols; carotenoids; cell walls; polymers; aldehydes; biosynthesis; biotechnology; geochemistry; literature; reviews
DNAL Call No.: 384 F773

121
Mechanism of Adaptation and Hydrogen Photoproduction in a Marine Green Alga Chlamydomonas-sp MGA 161
Miyamoto, K.; Nawa, Y.; Matsuoka, S.; Ohta, S.; Miura, Y.
Source: JOURNAL OF FERMENTATION AND BIOENGINEERING 70(1):66-69 (1990).
Descriptors: microorganism; fermentation; biotechnology industry
DNAL Call No.: QP601.A1J6
Abstract:

The adaptation process in a marine green alga, Chlamydomonas sp. MGA 161 was studied together with its light-dependent hydrogen evolution in terms of photosystem involvement and electron donation, and these processes were compared with those in Chlamydomonas reinhardtii. Hydrogen production in the light by MGA 161 was only a little more than that in the dark. Hydrogen metabolism in the illuminated cells of MGA 161 depended not on water but on cellular starch for as electron source.

122

Construction of Multiple Herbicide Resistant Ammonia Excreting Strains of Cyanobacterium Nostoc muscorum

Modi, D.R.; Singh, D.R.; Rao, A.K.; Chakravarty, K.S.; Singh, H.N.

Source: BIOTECHNOLOGY LETTERS 13(11):793-798 (1991).

Descriptors: nostoc muscorum; strains; gloeocapsa; herbicides; herbicide resistance; phenotypes; dna; genetic transformation; gene transfer; mutations; ammonia; excretion; photosystem ii; nitrogen fixation

DNAL Call No.: QR53.B56

Abstract:

Machete resistant (Matr), basalin resistant (Basr), 3(3,4 dichlorophenyl)-1,1-dimethyl urea resistant (DCMUr), atrazine resistant (Atr(r)) and propanil resistant (Prpr) phenotypes Gloeocapsa sp. were contransformed to Nostoc muscorum at high frequency. Spontaneously occurring mutants of the multiple herbicide resistant transformant containing L-methionine-DL-sulfoximine resistant (Msxr), ethylene diamine resistant (Edar) of phosphinothricin resistant (Pptr) glutamine synthetase (GS) showed extracellular liberation of ammonia resulting from fixation of N2 under photosynthetic conditions. Results suggest a definite role of GS activity in regulation of extracellular ammonia.

123

European Bioconversion Projects and Realizations for Macroalgal Biomass St.-Case-La Guildo France Experiment

Morand, P.; Charlier, R.H.; Maze, J.

Source: HYDROBIOLOGIA 204-205(0):301-308 (1989).

Descriptors: laminaria ulva; biomass utilization; compost biotechnology

DNAL Call No.: 410 H992

124

Storm Wrack of Marine Algae and Grasses as Raw Material for Bioconversion

Mun, T.H.; Kondrat'eva, L.M.; Garetova, L.A.

Source: SOVIET BIOTECHNOLOGY (1):68-70 (1989).

Descriptors: marine areas; algae; grasses; biomass production; raw materials; conversion; biotechnology

DNAL Call No.: TP248.13.S68

125

The Application of Two-Phase Aqueous Systems to the Purification of Phycoerythrin from Synechococcus-sp DC-2

Niven, G.W.; Smith, S.J.; Andrews, A.T.

Source: BIOTECHNOLOGY TECHNIQUES 4(6):373-378 (1990).

Descriptors: algae; marine; cyanobacterium; biotechnology industry

DNAL Call No.: TP248.24.B55

Abstract:

The potential of aqueous two-phase systems for the purification of phycoerythrin from a

marine cyanobacterium was investigated. Purities in excess of 90% total soluble protein were obtained in a single step processes and separation of two polymeric forms of phycoerythrin was achieved.

126
Market Applications for Microalgae
Rattray, J.B.M.
Source: JOURNAL OF THE AMERICAN OIL CHEMISTS' SOCIETY 66(5):648-653 (1989).
Descriptors: biotechnology industry; fats and oils
DNAL Call No.: 307.8 J82

127
Microbial Production of Glycerol and other Polyols
Rehm, H.J.
Source: BIOTECHNOLOGY: A COMPREHENSIVE TREATISE: BIOTECHNOLOGY, VOL. 6B. SPECIAL MICROBIAL PROCESSES. Rehm, H.J., editor. VCH Publishers, Inc., New York, 1989, pp.51-70.
Descriptors: review; yeast; bacteria; algae; biotechnology
DNAL Call No.: QR53.B52

128
Coupling of Solar Energy to Hydrogen Peroxide Production in the Cyanobacterium Anacystis-nidulans
Roncel, M.; Navarro, J.A.; De La Rosa, M.A.
Source: APPLIED AND ENVIRONMENTAL MICROBIOLOGY 55(2):483-487 (1989).
Descriptors: anacystis nidulans; hydrogen peroxide; biosynthesis; solar energy; photosynthesis; immobilization
DNAL Call No.: 448.3 AP5
Abstract:

Hydrogen peroxide production by the blue-green algae (cyanobacteria) under photoautotrophic conditions is of great interest as a model system for the bioconversion of solar energy. Our experimental system was based on the photosynthetic reduction of molecular oxygen with electrons from water by Anacystis nidulans 1402-1 as the biophotocatalyst and methyl viologen as a redox intermediate. It has been demonstrated that the metabolic conditions of the algae in their different growth stages strongly influence the capacity for hydrogen peroxide photoproduction, and so the initial formation rate and net peroxide yield became maximum in the mid-log phase of growth. The overall process can be optimized in the presence of certain metabolic inhibitors such as iodoacetamide and p-hydroxymercuribenzoate, as well as by permeabilization of the cellular membrane after drastic temperature changes and by immobilization of the cells in inert supports such as agar and alginate.

129
Potential Production of Protoplasts from Porphyridium-sp Using an Enzymatic Extract of its Predator Gymnodinium-sp
Roth-Bejerano, N.; Van Moppes, D.; Sivan, A.; Arad, S.
Source: BIORESOURCE TECHNOLOGY 38(2-3):127-132 (1991).
Descriptors: algae; species; genetic engineering; protoplasts; production; genetic improvement; gymnodinium; enzymes; extracts; cell walls; degradation; osmotic pretreatment; respiration rate; photosynthesis; growth curve; cell division
DNAL Call No.: TD930.A32
Abstract:

Production of biochemicals from red algae will become an agro-industrial reality only after

improvement of strain through genetic manipulation has been achieved. In the absence of sexual reproduction, preparation of protoplasts is a pre-requisite for genetic improvement of new strains. Although preparation of protoplasts from plant cells is a common technique, its application in red algae was limited. The unicellular alga Porphyridium sp. is encapsulated in a sulfated polysaccharide, the structure of which is still not fully known. A crude extract of a dinoflagelate Gymnodinium, a natural predator of Porphyridium cells in open cultures, was found to degrade Poryphyridium sp. polysaccharide enzymatically. Porphyridium cells treated with the crude Gymnodinium extract were exposed to various osmotic media (0-1.5 M sucrose), and their volume was measured. Volume increase was observed in diluted sucrose solutions up to 0.175 M. While further dilution of the external osmoticum to 0.1 M had little effect, dilution to 0.0 M (distilled water) led to cell rupture. Elevated concentrations of external osmoticum resulted in shrinkage of the treated cells. Such osmotic behavior indicates exposure of the cells and thus cleavage of the capsule. The treatment did not affect the viability of the cells, as evidenced by fluorescein diacetate (FDA) fluorescence, nor did it affect the respiration rate, but it did lower the photosynthetic rate to some extent. The growth curves for the treated cells exhibited a longer lag time than in the non-treated controls. Lowering the NaCl content in the growth medium resulted in a further increase in the lag time of the treated cells. These results indicate that the treatment lowers the ability of Porphyridium cells to divide. Ability to divide is eventually recovered with time, the recovery apparently depending upon the external osmoticum. The results indicate that Gymnodinium crude extract degrades Porphyridium cell wall and thus can be used for protoplast production.

130

Ammonium Photoproduction by Free and Immobilized Cells of Chlamydomonas-reinhardtii

Santos-Rosa, F. and Galvan, F.

Source: APPLIED MICROBIOLOGY AND BIOTECHNOLOGY 31(1):55-58 (1989).

Descriptors: calcium alginate-entrapped cells; chlorophyll membrane permeability; cell-matrix interaction; fermentation; biotechnology industry

DNAL Call No.: QR1.E9

Abstract:

Free-living or immobilized Chlamydomonas reinhardtii cells photoproduce ammonium from nitrite in a medium containing 1 mM of L-methionine-D,L-sulphoximine(MSX). Ammonium is accumulated in the medium to 8 mM final concentration, which inhibits nitrite uptake by the MSX-treated cells and consequently the excretion of ammonium is blocked. However, if ammonium was removed from the medium and nitrite and MSX periodically restored, the photoproduction process could be maintained over 96 h, with a final ammonium concentration of about 18 mM for free-living cells and 28 mM for immobilized ones. The MSX-treated cells showed a photoproduction productivity of 1300 .mu.mol NH4+ .cntdot. mg chlorophyll (Chl)-1, with an average production rate of 14 .mu.mol NH4+ .cntdot.mg chlorophyll Chl-1 per hour, for calcium alginate-entrapped cells, while the corresponding data for free-living ones was 650 .mu.mol NH4+ .cntdot. mg Chl-1 and 6.7 .mu.mol NH4 .cntdot. mg Chl-1 per hour, respectively. Immobilized cells showed a significant increase in the nitrite uptake rate, probably due to a change in membrane permeability as a consequence of cell-matrix interactions.

131

Biological Viability of Chlamydomonas-reinhardtii Cells Entrapped in Alginate Beads for Ammonium Photoproduction

Santos-Rosa, F.; Galvan, F.; Vega, J.M.

Source: JOURNAL OF BIOTECHNOLOGY 9(3):209-220 (1989).

Descriptors: photosynthesis; biotechnology

DNAL Call No.:
Abstract:

The green alga Chlamydomonas reinhardtii was immobilized by entrapment in Ca2+-alginate gel for ammonium photoproduction. The physical characteristics of the beads, their stability and ammonium-photoproduction capacity were affected by a variety of interactive factors, including cell loading, alginate viscosity and concentration, and the nature of the buffered medium. Electron micrographs show alterations in the morphology of the immobilized cells. Entrapped C. reinhardtii cells retained their biological viability, maintaining normal photosynthetic and respiratory activities, and they grew inside the gel beads, with a generation time of 9 h as compared with the 8 h shown by free cells under similar conditions. The unusually high rates for nitrite uptake and ammonium photoproduction, as well as the nitrite reductase activity shown by alginate-immobilized cells, were related to the changes in the membrane permeability induced by cell-matrix interaction.

132

Photoproduction of Ammonium by Chlamydomonas-reinhardtii Cells Immobilized in Barium Alginate a Reactor Feasibility Study

Santos-Rosa, F.; Galvan, F.; Vega, J.M.

Source: APPLIED MICROBIOLOGY AND BIOTECHNOLOGY 32(3):285-290 (1989).

Descriptors: chlamydomonas reinhardtii; ammonia; immobilization; barium; alginates

DNAL Call No.: QR1.E9

Abstract:

Chlamydomonas reinhardtii cells immobilized in Ba-alginate beads provide a stable and effective system for photoproducing ammonium from nitrite in a culture medium containing L-methionine-D,L-sulphoximine. The process was studied in either air-lift, fluidized- or packed-bed reactors, the last one providing the most suitable system with a volumetric activity of 2700 .mu.mol NH4+.cntdot.1-1 per hour.

133

Pharmaceuticals from Cultured Algae

Schwartz, R.E.; Hirsch, C.F.; Sesin, D.F.; Flor, J.E.; Chartrain, M.; Fromtling, R.E.; Harris, G.H.; Salvatore, M.J.; Liesch, J.M.; Yudin, K.

Source: JOURNAL OF INDUSTRIAL MICROBIOLOGY 5(2-3):113-124 (1990).

Descriptors: axenic; cyanobacteria; bacteria; microorganism; pachydictyol caulerpenyne haplindoles; antifungal depsipeptide; biotechnology industry; fermentation

DNAL Call No.: QR53.J68

Abstract:

An algae screening program, including cultured macroalgae, cultured cyanobacteria and cultured eukaryotic microalgae has been undertaken. Methods for the isolation, purification, preservation and cultivation of axenic cyanobacteria and eukaryotic cultures have been developed. Screening of these groups for biologically active components has lead to the isolation of pachydictyol and caulerpenyne from cultured macroalgae, while a series of hapalindoles and an antifungal depsipeptide have been isolated from cyanobacteria.

134

Applications of Some Algal Polysaccharides in Biotechnology

Skjak-Braek, G. and Martinsen, A.

Source: SEAWEED RESOURCES IN EUROPE: USES AND POTENTIAL. Guiry, M.D. and Blunden, G., editors. John Wiley and Sons, Inc., Somerset, New Jersey, 1991, 432 pp.

Descriptors: marine; cell culture medium; catalyst; immobilization; food technology; biomedics

DNAL Call No.: SH390.7S44

135
The Novel Non-Heme Vanadium Bromoperoxidase from Marine Algae Phosphate Inactivation
Soedjak, H.S.; Everett, R.R.; Butler, A.
Source: JOURNAL OF INDUSTRIAL MICROBIOLOGY 8(1):37-44 (1991).
Descriptors: industrial; enzyme stability; biotechnology industry
DNAL Call No.: QR53.J68
Abstract:

Vanadium bromoperoxidase is a naturally occurring vanadium-containing enzyme isolated from marine algae. V-BrPO catalyzes the oxidation of halides by hydrogen peroxide which can result in the halogenation of organic substrates. Bromoperoxidase activity is measured by the halogenation of monochlorodimedone (2-chloro-5,5-dimethyl-1,3-dimedone, MCD). In the absence of an organic substrate, V-BrPO catalyzes the halide-assisted disproportionation of hydrogen peroxide yielding dioxygen. The dioxygen formed is in the single excited state (1O2). V-BrPO is quite stable to thermal denaturation and denaturation by certain organic solvents which makes V-BrPO an excellent candidate for industrial applications. The stability of V-BrPO in the presence of strong oxidants and in the presence of phosphate is reported. Incubation of V-BrPO in phosphate buffer (1-100 mM at pH 6; 2-10 mM at pH 5) inactivates the enzyme. The inactivity can be fully restored by the addition of vanadate if excess phosphate is removed. The inactivation of V-BrPO by phosphate can be prevented by the presence of H202 (4-40 mM). We are currently investigating the mechanism of V-BrPO inactivation by phosphate. V-BrPO was not inactivated by HOCl (1 mM) nor H202. In addition V-BrPO was not inactivated under turnover conditions of 1 mM H202 with 0.1-1 M Cl- at pH 5 nor 2 mM H202 with 0.1 M Br-.

136
Environmental Control of Lipids and Fatty Acid Production in the Diatom Navicula-saprophila
Sriharan, S.; Bagga, D.; Sriharan, T.P.
Source: APPLIED BIOCHEMISTRY AND BIOTECHNOLOGY 20-21(0):281-292 (1989).
Descriptors: navicula; lipids; fatty acids; production; temperature relations; biomass; renewable resources
DNAL Call No.: QD415.A1J62

137
Biological Active Substances from Algae
Taeymans, D.
Source: MEDED FAC LANDBOUWWET RIJKSUNIV GENT 54 (4B):1597-1602 (1989).
Descriptors: phycocolloids; enzymes; vitamins; amino acids; coloring agents; single cell protein; single cell lipid; biotechnology

138
Glycolate Photoproduction by Free and Alginate-Entrapped Cells of Chlamydomonas-reinhardtii
Vilchez C.; Galvan, F.; Vega, J.M.
Source: APPLIED MICROBIOLOGY AND BIOTECHNOLOGY 35(6):716-719 (1991).
Descriptors: algae; immobilization; biotechnology industry
DNAL Call No.: QR1.E9
Abstract:

Chlamydomonas reinhardtii cells provide an effective system for glycolate photoproduction, operative during 30 h when they are growing under low CO2, in the presence of 1 mM aminooxyacetate and 50 .mu.M acetazolamide. Glycolate excretion by

the cells can proceed for about 4 days if every other 12 h a high CO2 level is restored in the culture in the absence of inhibitors. The immobilized system in alginate beads has about a twofold higher glycolate photoproduction rate (23 .mu.mol .cntdot. mg chlorophyll (Chl)-1 .cntdot. h-1) than free-living cells (12 .mu.mol .cntdot. mg Chl-1 .cntdot. h-1).

139

Production of L Leucine from Alpha Ketoisocaproic Acid by Cell-Free Extract of Euglena-gracilis Z
Yoshimura, T.; Koike, N.; Kimura, Y.; Yamaoka, R.; Hayashiya, K.
Source: JOURNAL OF FERMENTATION AND BIOENGINEERING 70(6):427-428 (1990).
Descriptors: algae; alpha ketoglutarate decarboxylase; biotechnology industry
DNAL Call No.: QP601.A1J6
Abstract:

L-Leucine was produced from .alpha.-ketoisocaproic acid at about 100% conversion with L-glutamate as an amino donor using cell-free extracts of Euglena gracilis Z. .alpha.-ketoglutarate decarboxylase in Euglena drives the conversion to completion by removal of .alpha.-ketoglutarate formed during the transamination.

140

Isolation and Analysis of Nitrate Reductase Deficient Mutants for use in Genetic Engineering of Microalgae for Increased Fuel Production
Zeiler, K.G. and Brown, L.M.
Source: JOURNAL OF PHYCOLOGY 27(3 suppl.):80 (1991).
Descriptors: abstract; monoraphidium-minutum; cyclotella-cryptica
DNAL Call No.: QK564.J6

Bioremediation Using Algae

141

A Bioseparation Process for Removing Lead-II Ions from Waste Water by Using Chlorella-vulgaris
Aksu, Z. and Kutsal, T.
Source: JOURNAL OF CHEMICAL TECHNOLOGY AND BIOTECHNOLOGY 52(1):109-118 (1991).
Descriptors: algae; microorganism; metals; wastewater treatment; biotechnology industry; batch reactor; bioreactor
DNAL Call No.: QR53.J685
Abstract:

Biosorption of heavy metals by microbial cells has been recognized as a potential alternative to existing technologies for removing heavy metals from industrial waste waters. Many aquatic microorganisms, such as algae, can take up dissolved metals from their surroundings to their cells. In this study, the adsorption of lead(II) ions was investigated in a single-staged batch reactor. Chlorella vulgaris, a green alga, was used as the sorbent. The sorption phenomenon was expressed by the Freundlich adsorption isotherm and this expression was used for the calculation of residual or adsorbed metal ion concentration at equilibrium (Ceq or Cx, eq) at a given 'volume of waste water containing heavy metal ion/quantity of alga (Vo/Xo)' ratio in a single-staged batch reactor. Experimental Ceq and Cx, eq values were compared to calculated ones. Applications in waste water treatment for lead (II) removal have been suggested.

142
Review of Biotechnology Applications to Nuclear Waste Treatment
Ashley, N.V. and Roach, D.J.W.
Source: JOURNAL OF CHEMICAL TECHNOLOGY AND BIOTECHNOLOGY 49(4):381-394 (1990).
Descriptors: review; algae; bacteria; fungus; metal; radionuclide biosorption; biopolymer; industrial waste treatment
DNAL Call No.: QR53.J685

143
Microbiotests in Aquatic Ecotoxicology Characteristics Utility and Prospects
Blaise, C.
Source: ENVIRON TOXICOL WATER QUAL 6(2):145-156 (1991).
Descriptors: bacteria; protozoa; microalgae; invertebrate; fish cell line; hazard assessment; biotechnology; immunochemistry; cost effectiveness
Abstract:

Small-scale biological tests (microbiotests) have steadily increased in development and application over the last 30 years in the field of aquatic ecotoxicology. Multitrophic level assessment requirements, attractive features of microbiotests, and the constant search for simplicity and cost efficiency of testing are reasons explaining the expanding use of microbiotests. In this article, the major characteristics that advantageously confer popularity on microbiotests are presented and 25 currently applied aquatic toxicity microbiotests are listed. Conducted with bacteria, protozoans, microalgae, small invertebrates, and fish cell lines, these microbiotests represent a realistic cross section of those that are now becoming an essential part of ecotoxicological assessment. Microbiotests can be profitably employed for ranking and screening chemicals, for novel applications enabling rapid detection of ecotoxic effects in complex liquid samples, and for increasing the cost efficiency and diagnostic potential of hazard assessment schemes. Microbiotesting research, development, and applications will continue to surge in the 1990s, driven, among other factors, by the imperative need for cost effectiveness in environmental programs. Research in the fields of ecotoxicology, biotechnology, and immunochemistry should provide interesting breakthroughs to further enhance the specificity and diagnostic value of microbiotests.

144
Selection of Surrogates for a Genetically Engineered Microorganism with Cellulolytic Capability for Ecological Studies in Streams
Bott, T.L. and Kaplan, L.A.
Source: CANADIAN JOURNAL OF MICROBIOLOGY 37(11):848-857 (1991).
Descriptors: cellulomonas-sp; cellulomonas-uda; cladophora-glomerata; liriodendron-tulipifera; genetic engineering; statistics; biodegradation; flowing-water microcosms
DNAL Call No.: 448.8 C162
Abstract:

Aerobic cellulolytic bacteria were ranked according to ability to degrade cellulose azure and to clear cellulose agar. Cellulomonas uda NRRL B404 and Cellulomonas sp. NRC 2406 showed greater clearing of cellulose agar than other isolates, but differences in cellulose azure decomposition were not statistically significant. Isolates were tested for ability to accelerate decomposition of tulip poplar (Liriodendron tulipifera) leaves and Cladophora glomerata (Chlorophyta) detritus in stream water. There was significantly more cellulose lost from leaves exposed to Cellulomonas flavigena NRC 2403, Cellulomonas fimi NRRL B402, Cellulomonas sp. NRC 2406, and Cellvibrio gilvus ATCC 13127 and NRC 2407 than in the stream-water control, and the weight losses of leaves

exposed to some isolates were significantly greater than in the control. There was significantly more cellulose lost from Cladophora glomerata detritus exposed to these and five other isolates, and there were greater weight losses than in the stream-water control. Cellulomonas uda NRRL B404 was the slowest growing isolate, although growth rates of isolates did not differ statistically. Cellulomonas uda NRRL B404, Cellulomonas sp. NRC 2406, Cellulomonas fimi NRRL B402, Celllulomonas flavigena NRC 2403, and Cellvibrio gilvus ATCC 13127 were selected as the best candidates for larger scale experiments. Persistence of Cellulomonas uda, Cellulomonas sp. NRC 2406, and Cellulomonas sp. CS1-1 in stream-bed sediments was studied in flowing-water microcosms, using fluorescent antibodies and epifluorescence microscopic counts to asses densities of target cells. Isolate densities declined from postinoculation maxima, but organisms were detected 2-4 weeks later in three different experiments.

145

Applied Microbial Processes for Metals Recovery and Removal from Wastewater
Brierley, C.L.; Brierley, J.A.; Davidson, M.S.
Source: METAL IONS AND BACTERIA. Beveridge, T.J. and Doyle, R.J., editors. John Wiley and Sons, Inc., Somerset, New Jersey, 1989, pp.359-382.
Descriptors: review; bacteria; fungi; algae; biotechnology
DNAL Call No.: QR92.M45M47

146

Options for the Rational Design and Operation of Oxidation Ponds
Carberry, J.B.
Source: WATER SCIENCE AND TECHNOLOGY 24(5):21-32 (1991).
Descriptors: incident sunlight; diurnal; ph variation; calcium chloride addition; wastewater treatment; algal bacterial clay treatment system; computer model; methods; biotechnology; microorganism

147

Method for Evaluating Algal Production and Degradation Based on Nitrogen Levels in Particulate Organic Matter
De Casabianca-Chassany,M.L.
Source: BIORESOURCE TECHNOLOGY 39(1):1-8 (1992).
Descriptors: ulva-rotundata; algae; free algal biomass; algal growth; carbon level; photosynthesis; coastal lagoon; water pollution; environmental quality; biotechnology industry
DNAL Call No.: TD930.A32
Abstract:

A population of Ulva rotundata (Bliding, C., A critical survey of European taxa in Ulvales. Part I. Opt. Bot. Lund, A8 (3), 1-160) was studied for 7 months in a shallow brackish eutrophic lagoon (Prevost Lagoon, Languedoc, France). Biomass was measured in square-meter quadrates, and algal growth in on-site cages. Carbon and nitrogen levels were spatiotemporally monitored in Ulva thalli and in particulate organic matter. Thalli rapidly degraded into particulate matter from the surface to the bottom, increasing nitrogen concentrations and decreasing carbon concentrations. We propose a method for estimating algal production and possible carbon loss in this type of lagoon, based on nitrogen levels in particulate organic matter. Algal production over the whole period can be evaluated by adding instantaneous biomass levels corresponding to the main particulate nitrogen peaks.

148

Accumulation of Metals by Microorganisms and Algae

Gadd, G.M.
Source: BIOTECHNOLOGY: A COMPREHENSIVE TREATISE: BIOTECHNOLOGY, VOL. 6B. SPECIAL MICROBIAL PROCESSES. Rehm, H.J., editor. VCH Publishers, New York, 1989, pp.401-434.
Descriptors: review; bacteria; algae; fungi; yeast; precipitation; binding complex formation recovery; transformation; biotechnology
DNAL Call No.:

149
Nutrient Removal from Secondary Effluent by Filamentous Algae
Hashimoto, S. and Furukawa, K.
Source: JOURNAL OF FERMENTATION AND BIOENGINEERING 67(1):62-69 (1989).
Descriptors: oscillatoria-sp; secondary activated sludge effluent; continuous culture; kinetic constants; wastewater treatment; biotechnology industry
DNAL Call No.: QP601.A1J6
Abstract:

The nutrient removal potential of filamentous Oscillatoria sp. was quantitatively studied. Oscillatoria sp. showed satisfactory growth on secondary activated sludge effluent supplemented with 1% NaHCO3. Continuous open culture of Oscillatoria sp. was kept stable using a continuous stirred tank equipped with a filter-separator. Kinetic constants YN and YP were 10.6 g cells/g NO3-N and 37.7 g cells/g PO43- respectively through the analysis of the results of continuous culture experiments. Monoalgal continuous culture of more than 90% purity could be maintained for 6 months without contamination. The harvested Oscillatoria cells were proven to have an excellent filterability. They also have excellent autoflotation. The amino acid composition of the Oscillatoria algal protein compares favorable with the tentative standard for ideal protein defined by FAO/WHO.

150
Removal of Lead from Contaminated Soil and Water Using a Mixed Microbial Ecosystem
Ibeanusi, V. and Archibold, E.
Source: ABSTR ANNU MEET AM SOC MICROBIOL 90(0):327 (1990).
Descriptors: abstract; bacteria; algae pond; biotechnology; bioremediation
DNAL Call No.: 448.39.S012A

151
Mechanisms of Mixed Microbial Mobilization and Recovery of Heavy Metals in a Simulated Pond System
Ibeanusi, B.; Archibold, E.; Bender, J.; Gould, J.
Source: ABSTR ANNU MEET AM SOC MICROBIOL 89(0):362 (1989).
DNAL Call No.: 448.39.S012A

152
Accumulation of Cobalt by Marine Alga
Kuyucak, N. and Volesky, B.
Source: BIOTECHNOLOGY AND BIOENGINEERING 33(7):809-814 (1989).
DNAL Call No.: 381 J8224

153
Desorption of Cobalt-Laden Algal Biosorbent
Kuyucak, N. and Volesky, B.
Source: BIOTECHNOLOGY AND BIOENGINEERING 33(7):815-822 (1989).
Descriptors: ascophyllum-nodosum accumulation; metals; calcium chloride solution; cellular

structure; biotechnology industry
DNAL Call No.: 381 J8224
Abstract:

Following an effective accumulation of cobalt by nonliving algal biomass of Ascophyllum nodosum, the desorption release of the metal from the biosorbent was examined, using H2SO4, HCl, NH4OH, KHCO3, EDTA, KSCN, KCl, and CaCl2 solutions. The solution of CaCl2 (0.05M) in HCl appeared to be the best eluant capable of desorbing more than 96% of the sequestered cobalt at the optimum pH 2-3. The optimum solid-to-liquid ratio was more than 10 with the cobalt reuptake capacity of the biosorbent undiminished. The effect of temperature on the elution process and the elution rate was not significant up to 60.degree.C. The infrared (IR) spectra of the native and the eluted biomass did not show significant differences. The electron micrographs of the algal biomass taken after washing it with the CaCl2 (0.1M) eluant solution indicated no damage to the cells and cell walls, while strong acid, alkaline, and KSCN treatment resulted in some changes in the cellular structure. The kinetics of the cobalt stripping process was quite rapid. The required contact time for the complete metal removal from the biomass was shorter than 2h, even for the highest level of cobalt initially deposited on the biomass.

154
The Mechanism of Cobalt Biosorption
Kuyucak, N. and Volesky, B.
Source: BIOTECHNOLOGY AND BIOENGINEERING 33(7):823-831 (1989).
Descriptors: ascophyllum-nodosum accumulation; cell wall binding metals; biotechnology industry
DNAL Call No.: 381 J8224
Abstract:

Nonliving biomass of the common seaweed Ascophyllum nodosum is capable of accumulating cobalt from aqueous solutions to the extent of 160 mg Co2+/g. Successful desorption of cobalt from the biomass by acidic CaCl2 solutions revealed that the metal uptake phenomenon is reversible, implying physical sorption of cobalt. Chemical and instrumental analysis, including electron microscopy, infrared (IR) spectroscopy, X-ray dispersion and diffraction analysis provided supporting evidence that the biosorption mechanism involves predominantly ion exchange. Alginates of the cell wall (-COOH groups) play an important role in cobalt binding. Coordination and sorption in the cell wall structure occur simultaneously and rapidly whereas penetration of cobalt into the cell occurs at a lower rate.

155
Sorption of Strontium-90 and Yttrium-90 by Anacystis Cells
Liu H-H; Chen, W-L; Wu, J-T.
Source: SCIENCE OF THE TOTAL ENVIRONMENT 91(0):275-282 (1990).
Descriptors: biosorbent; biotechnology; respiration; passive adsorption; light vs. dark acid desorption
Abstract:

A study of the sorption and desorption of strontium-90 and yttrium-90 by Anacystis cells showed a concentration factor of 4 .times. 104 ml g-1. The uptake of both radionuclides by the cells was very rapid and a linear relationship exists between the amount of radionuclides accumulated in the cells and those in the surrounding medium. The results show that the uptake of Sr/Y by the cells is primarily by passive adsorption rather than active absorption. There was no significant difference in the sorption rate between cells incubated in the light and those left in the dark. However, both heat treatment and the addition of a respiration inhibitor reduced the sorption of radionuclides by up to 10%. The radionuclides accumulated in the cell and could readily be desorbed by washing with

acid. Thus, this organism can be used as a biosorbent for concentrating and removal of these two radionuclides.

156

Bioremoval of Copper and Uranium from Solution Using Alginate-Immobilized Microcystis-spp
Lopez, S.L.; Pryfogle, P.A.; Stoner, D.L.; Dugan, P.R.
Source: ABSTR ANNU MEET AM SOC MICROBIOL 90(0):327 (1990).
Descriptors: abstract; biotechnology
DNAL Call No.: 448.39.SO12A

157

Evaluation of the Metal Uptake of Several Algae Strains in a Multicomponent Matrix Utilizing Inductively Coupled Plasma Emission Spectrometry
Mahan, C.A.; Majidi, V.; Holcombe, J.A.
Source: ANALYTICAL CHEMISTRY 61(6):624-627 (1989).
Descriptors: chlorella-pyrenoidosa; stichococcus-bacillaris; chlamydomonas-reinhardti; blue-green algae; iron; lead; copper; analytical preconcentration technique; biotechnology industry
DNAL Call No.: 381 J825A
Abstract:

Three freshwater heat-killed, lyophilized blue-green algae strains have been characterized as to their ability to accumulate heavy metals with a focus on the utilization of these algae as an analytical preconcentration technique. This study examines the metal uptake in several multicomponent mixtures by using inductively coupled plasma optical emission spectrometry (ICP-OES). Six milligrams of a pure strain of algae was added to 20-mL aliquots of buffered (ph 5.5-6.5) multielement solutions containing 0.1, 0.5, 1.0, 2.0, and 4.0 mg/L of K, Mg, Ca, Fe, Sr, Co, Cu, Mn, Ni, V, Zn, As, Cd, Mo, Pb, and Se. All three algae strains exhibit relatively high adsorption affinities for Fe, Pb, and Cu, with uptake between 70 and 98% at the 4 ppm concentration level. Biosorption occurs for essentially every element with the relative affinities decreasing in the order Pb > Fe > Cu > Cd > Zn > Mn > Mo > Sr > Ni > V > Se > As > Co for Chlorella pyrenoidosa at the 4 mg/L concentration level. Although some minor differences were seen, the other algae strains (Stichococcus bacillaris and Chlamydomonas reinharti) displayed similar adsorption behavior over the concentration range studied, indicating similar cell wall binding sites. Langmuirian isotherms exhibited a minimum of two slopes over the concentration range of 0.1-4.0 mg/L, indicating the probable existence of a least two adsorption mechanisms.

158

Retardation of Toxic Heavy Metal Dispersion from Nickel-Copper Mine Tailing Sudbury District Ontario Canada Role of Acidophilic Microorganisms II. Structure and Microanalysis of Bioprecipitants
Mann, H.; Tazaki, K.; Fyfe, W.S.: Wiseman, M.
Source: BIORECOVERY 1(3):173-188 (1989).
Descriptors: algae; carbon; oxygen; iron; sulfur mineralization; cellular deposits; biotechnology
DNAL Call No.: TA418.74B56
Abstract:

Fe-rich sediments in the tailings effluent of the Sudbury Ni, Cu mining area, northern Ontario, are predominantly magnetite and ferrihydrite, with subordinate akaganeite (.gamma.-FeOOH), goethite (.alpha.-FeOOH), hematite (.alpha.-Fe2O3) and vivianite [Fe3(PO4)2.8H2O]. The iron is derived from biologically mediated oxidative breakdown of Fe-sulphide minerals in the mine tailings waste, which generates acidic effluent with enhanced concentrations of aqueous Fe and SO42-. Surface analysis of the sediments by ESCA reveals that C (57 atom %), O (31%) and Fe (6.7%) are most abundant, in accord

with the presence of ubiquitous acidophilic microorganisms and their remnants, and Fe-oxide and oxyhydroxide minerals. The high resolution spectra of S (S2p) indicates partitioning of S between SH (163.4 e V), SO3 (167.3 ev) and SO42- oxidation states, with SO42- the most abundant, as the mineral gypsum (CaSO4). Similarly, carbon (C1s spectrum) is partitioned between hydrocarbon (285 eV) and carbonate (289.2 eV); and iron (Fe2p spectrum) between FeS, Fe(C5H5)(CO)3, FeSO42- and FeOOH. Both C and Fe spectra show a relationship between organic and inorganic compounds, suggesting that Fe-mineralization is linked to the microorganisms. TEM electron micrographs revealed ubiquitous remains of microorganisms in the sediments, heavily encrusted with Fe-oxide and oxyhydroxide minerals. Magnetite, maghemite, ferrihydrite, and geothite associated with cell walls, and intracellular sites, were identified from d-spacings of electron diffraction patterns. Nucleation of iron oxide minerals by acidophilic microorganisms appears to be ubiquitous in acidic Fe-rich tailings environments. The precise synthetic pathways of Fe mineral formation are not known.

159

Retardation of Toxic Heavy Metal Dispersion from Nickel-Copper Mine Tailing Sudbury District Ontario Canada Role of Acidophilic Microorganisms I. Biological Pathway of Metal Retardation

Mann, H.; Fyfe, W.S.; Kerrich, R.; Wiseman, M.

Source: BIORECOVERY 1(3):155-172 (1989).

Descriptors: euglena; iron; molybdenum; thorium; aluminum; zinc; manganese; cadmium; titanium; sequestration; lake; biotechnology

DNAL Call No.:

Abstract:

Leaching of .apprx. 600 M tonnes of mine tailings waste from the Sudbury nickel-copper mining district, Ontario, by infiltration of precipitation and dewatering of tailings, results in the release of .apprx. 41,000 kg of Ni and other toxic heavy metals to the environment per year. Tailings effluent waters are variably acidic (pH 3.5-6.3) due to the generation of H2SO4 by the biologically-mediated oxidation of sulphide minerals in the tailings, and contain enhanced levels of Fe (200x), Ni (8300x), Cu (670x), Mo (240x) and Th (1,300x) relative to concentrations in world average river water. Acidophilic microorganisms (Euglena sp.) thrive in the low pH, heavy metal laden discharge, and act as efficient scavengers of the aqueous solutes, such that bulk samples of algae are characterized by abundances of Fe, Al, Zn, Mn, Cd, Ti and Ni 103 to 105 times the aqueous solute concentration (by dry weight). Where tailings discharge is treated with "limestone slurry" to raise the pH, organic-rich sediments in lakes are enriched in Ni, Cu and other heavy metals, endorsing the role of microorganisms in sequestering toxic heavy metals from solutions, thereby diminishing the metal loading, and retarding their dispersion into the natural environment.

160

A Photo-Bioreactor Using Algal Phototaxis for Solids-Liquid Separation

Nakajima, T. and Takahashi, M.

Source: WATER RESEARCH 25(10):1243-1248 (1991).

Descriptors: euglena-gracilis; algae culture medium; wastewater treatment; bioengineering

DNAL Call No.: TD420.W3

Abstract:

Using algal-positive photoaxis, the possibility of keeping a high density of algae and separating them from water (i.e. solids-liquid separation) for removal of nutritive substances was investigated. Euglena gracilis shows positive phototaxis. Culture medium containing the alga in a culture vessel (bioreactor) was transferred to a shaded vessel (i.e., photo-clarifier), part of which was exposed to a spotlight. The organisms gathering

around the light were returned to the culture vessel, and the effluent was taken out from the shaded part and discharged out of the system. Two types of bioreactors having different types of photo-clarifiers were employed: vertical form (Type A) and horizontal form (Type B). Densities of E. gracilis in the culture vessel and effluent were examined in both types over the experimental period, and compared with a control system without a photo-clarifier. In both types the E. gracilis density in the effluent was lower than that in the culture vessel, whereas in the control system the density in the effluent was almost identical with that in the culture vessel. Separation efficiency was higher in Type B than in Type A. The obtained results indicate that it is possible to achieve solids-liquid separation by using algal phototaxis, and suggest a possibility of further improvement in the separation efficiency by modifying the structure of the photo-clarifier.

161
Comparative Water Quality Dynamics in a Recirculating Systems with Solids Removal and Fixed-Film or Algal Biofiltration
Rakocy, J.E.; Hargreaves, J.A.; Bailey, D.S.
Source: JOURNAL OF THE WORLD AQUACULTURE SOCIETY 22(3):49A (1991).
Descriptors: abstract; oreochromis-niloticus; bacteria; biotechnology
DNAL Call No.: SH138.W62

162
Phosphorus Uptake Kinetics of Immobilized Chlorella in Batch and Continuous-Flow Culture
Robinson, P.K.; Reeve, J.O.; Goulding, K.H.
Source: ENZYME AND MICROBIAL TECHNOLOGY 11(9):590-596 (1989).
Descriptors: biotechnology; waste disposal reactor; stocking density; process optimization
DNAL Call No.: TP248.E5E565
Abstract:

Study has been made of the uptake of orthophosphate phosphorus (PO4-P) by Chlorella emersonii (CCAP 211/8a) entrapped in 4-mm diameter Ca-alginate beads. In batch culture studies, 100 beads, each stocked with 107 cells, removed all PO4-P from 100 ml synthetic growth medium (i.e. about 10 .mu.mol) in under 24 h. Uptake followed exponential kinetics with medium PO4-P concentration falling by 50% every 2.0 h, when external concentrations were above 6 .mu.M. A stocking density of 107 cell/bead (the highest tested) was found to be suitable for rapid phosphorus removal, though this was not necessarily optimal with respect to cellular efficiency. Small-scale packed-bed reactors containing 10 ml gel were able to remove up to 240 .mu.mol PO4-P from 4-5 l medium over 10-12 day experimental periods. Uptake efficiencies ranged from 29 to 97% and averaged 66% and 44% with synthetic growth medium and secondary treated effluent, respectively. Uptake kinetics are analyzed and results discussed with respect to process optimization.

163
The Engineering of Microalgae Mass Cultures for Treatment of Agricultural Wastewater, with Special Emphasis on Selenium Removal from Drainage Waters
Shelef, G.
Source: BIOTREATMENT OF AGRICULTURAL WASTEWATER. Huntley, Mark E., editor. CRC Press, Boca Raton, FL, 1989, pp.143-148.
Descriptors: environmental pollution; agricultural wastes; drainage water; pollutants; selenium; waste water treatment; biotechnology; algae
DNAL Call No.: TD755.B48

164

An Aerobic Piggery Slurry Treatment System with Integrated Heat Recovery and High-Rate Algal Ponds

Svoboda, I.F. and Fallowfield, H.J.

Source: WATER SCIENCE TECHNOLOGY 21(4-5):277-288 (1989).

Descriptors: biotechnology industry; metabolic heat; mathematical model; continuous culture reactor; livestock industry; agriculture; wastewater treatment; sewage disposal

165

Waste Water Treatment Using Saline Cultures of Microalgae

Toha, J.; Soto, M.A.; Cuadros, X.

Source: BIOTECHNOLOGY TECHNIQUES 4(6):441-444 (1990).

Descriptors: aphanothece-sp; dunaliella-sp; escherichia-coli; microorganism; algae; bacteria; photosynthesis; stabilization pond; wastewater treatment; biotechnology industry

DNAL Call No.: TP248.24.B65

Abstract:

Survival of microorganisms (Escherichia coli has been used as an example) is affected by a combination of salinity and high pH induced by the active photosynthesis of marine microalgae (Aphanotece or Dunaliella sp.). This effect can be applied to create a more efficient wastewater treatment process using algal stabilization ponds.

166

Mercury Accumulation and Volatilization in Immobilized Algal Cell Systems

Wilkinson, S.C.; Goulding, K.H.; Robinson, P.K.

Source: BIOTECHNOLOGY LETTERS 11(12):861-864 (1989).

Descriptors: chlorella; algae; wastewater treatment methods; biotechnology

DNAL Call No.: QR53 B56

Abstract:

Rapid removal of mercury from growth medium and its uptake by free and alginate-entrapped Chlorella has been observed. Immobilized cell systems accumulated more mercury than free cell systems. In addition, both volatilized significant quantities of mercury. Studies show, however, that mercury lost in this way may re-enter the aqueous phase and subsequently be accumulated by immobilized cells.

167

Removal of Organochlorine Compounds in an Upflow Flocculated Algae Photobioreactor

Wu, X. and Kosaric, N.

Source: WATER SCIENCE TECHNOLOGY 24(5):221-232 (1991).

Descriptors: chlorella scenedesmus; biodegradation; chlorobenzene 2 4 dichlorophenol; wastewater treatment; biotechnology industry; bioreactor

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